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THE ROWPU PREFILTRATION SYSTEM: REMOVAL OF MICROORGANISMS

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The Army has developed a Reverse Osmosis Water Purification Unit (ROWPU) to provide potable water during field operations. The unit uses coagulation, multimedia filtration, cartridge filtration, RO, and chlorination to provide a potable product from a variety of contaminated water sources.

The objective of this study was to evaluate the ability of the treatment system, minus reverse osmosis and chlorination, to remove microorganisms from

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fresh water under various conditions of water quality and at various system loading rates. If such a capability could be demonstrated, higher production rates could be realized at a lower energy cost.

The system was assembled to process Monocacy River water. Bacillus globigii and Poliovirus I, LSc strain, were inoculated into river water. These organisms were enumerated at different locations within the system, as were naturally-occurring Escherichia coli, and the total aerobic and enteric bacterial groups. Nine-hour production tests were performed at 30, 35, and 40 gallon/minute flow rates. Nine such tests were performed in Sep-Oct 1980.

Microbiological enumerations indicated the following mean percent removals of microorganisms in the prefiltration system: Bacillus globigii, 98.3; Escherichia coli, 93.5; Poliovirus I, LSc strain, 80.3; total aerobic bacteria, 83.1; and total enteric bacteria, 86.8. Influent and process waters were also assayed for pH, alkalinity, total dissolved solids, total organic carbon, and turbidity.

The multimedia filter was the major unit effecting removal. Implications of the results with respect to field operations are presented.

PREFACE

The authors wish to acknowledge the interest and assistance of the following personnel in the formulation of test plans, the setup of equipment, and the conduct of experiments: from US Army Mobility Equipment Research and Development Command (USAMERADCOM), Messrs. Donald Lindsten, Harry Goto, Peter Pedersen, and Roger Anzzolin; and from US Army Medical Bioengineering Research and Development Laboratory (USAMBRDL), Messrs. William Rose, and Ralph Chyrek.

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TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

INTRODUCTION

In 1979 the Army type-classified a 600-gallon per hour reverse osmosis water purification unit (ROWPU) to provide a water treatment capability during field operations.[1] The 600-gallon ROWPU was envisioned as the first of a family of water purification units to be a replacement for the Erdlator, in use since World War II. As the name indicates, reverse osmosis (RO) is the primary unit process in the ROWPU treatment system. Other unit processes, multimedia filtration and cartridge filtration, are included for conditioning the water prior to reverse osmosis treatment. Hypochlorination is included for disinfection following reverse osmosis treatment. The ROWPU unit, in addition to providing treatment of fresh water, is also highly effective in reducing dissolved solids from brackish or sea water to a potable level. Also, the ROWPU will provide some treatment of unconventional warfare pollutants with minimal add-on equipment.[2]

Most fresh water sources do not require treatment for reducing or removing dissolved solids. For such sources, use of the entire ROWPU treatment train may be unnecessary. More specifically, treatment through the multimedia and cartridge filters, coupled with postchlorination, may suffice to provide potable water. Two potential results of the elimination of the reverse osmosis process from the treatment system are noteworthy. First, the production rate could be increased to approximately 1,800 gallons per hour, the design capacity of the prefiltration system. Second, approximately 50 percent of the power used in the 600-gallon per hour ROWPU is used to pressurize and pump water through the RO membranes. Therefore, if water could be processed for drinking without use of the reverse osmosis system, a significant fuel savings could be realized.

A formal decision by the US Army to use the prefiltration system (multimedia filter - cartridge filter treatment train) with postchlorination for the treatment of fresh water hinges on the demonstration of the ability of the system to provide a potable water product. Although the capability of the prefiltration system to remove suspended solid particles has been demonstrated, there is some question concerning the ability of the system to remove microorganisms. The effects of chlorination on these organisms are well documented;[3] however, the capability of the prefiltration system to remove such organisms prior to chlorination is not as well defined.

In 1974, Ford and Pressman[4] studied the performance of a prototype ROWPU filter rated at 360 gallons per hour. Raw river water was seeded with f2 coliphage virus prior to filtration. The virus and indigenous coliform bacteria were assayed in samples collected throughout the ROWPU treatment train. The multimedia filter used was described as a "3-inch layer of graded gravel supporting a 9-inch layer of sand and a 15-inch layer of coal." The filter was loaded at 10 gallons per minute per square foot. Several runs of short duration were made during the study, representative results of which are summarized in Table 1.

The design of the filtration system has changed significantly since this prototype. To describe adequately the removal of microorganisms by the prefiltration system, additional data were required. The objective of this study was to evaluate the ability of the 600-gallon per hour ROWPU prefiltration

system to remove microorganisms from untreated fresh water under different water quality conditions and flow rates.

TABLE 1. REPRESENTATIVE MULTIMEDIA FILTER REMOVAL DATA OF PROTOTYPE ROWPU[4]

Run	Time (min)[b]	Percent Removal[a]		
		Total Coliform	Fecal Coliform	f2 Coliphage
2	30-60	78.4	91.1	99.999
	90-120	0	98.4	99.994
	150-180	79	91.8	99.998
	210-240	97.1	95.9	99.999
3	30 (Grab)	96.2	--	98.1
	60-120	96.4	--	99.9
	120-180	96	98.7	99.8

a. Coliform assays by fermentation tube, results based on most probable number.
b. Unless stated, composited sample.

MATERIALS AND METHODS

SITE LOCATION AND EXPERIMENTAL APPARATUS

The test site was on Fort Detrick property, located adjacent to the Monocacy River on the grounds of the Fort Detrick Sewage Treatment Plant. A description of the test site area is presented in Figure 1. The raw water intake for the test apparatus was placed upstream of the sewage treatment plant outfall. The test apparatus utilized in conducting these experiments is described in Figure 2. Water was pumped from the river via the centrifugal pumps (see Appendix A for details on these pumps and other equipment). Prior to polymer addition, non-indigenous microorganisms were seeded into influent river water. Table 2 lists the microorganisms used in this study.

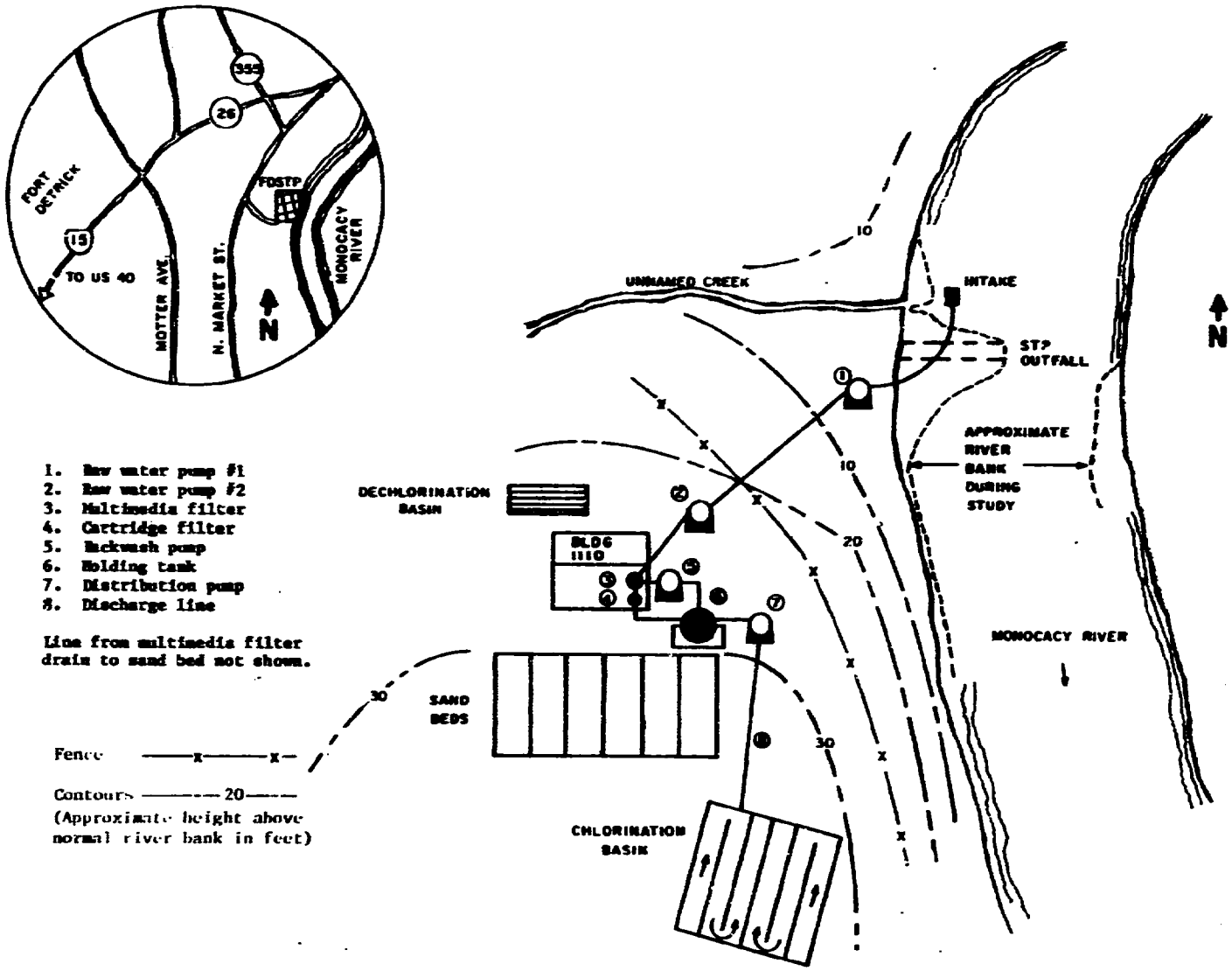


Figure 1. Test site.

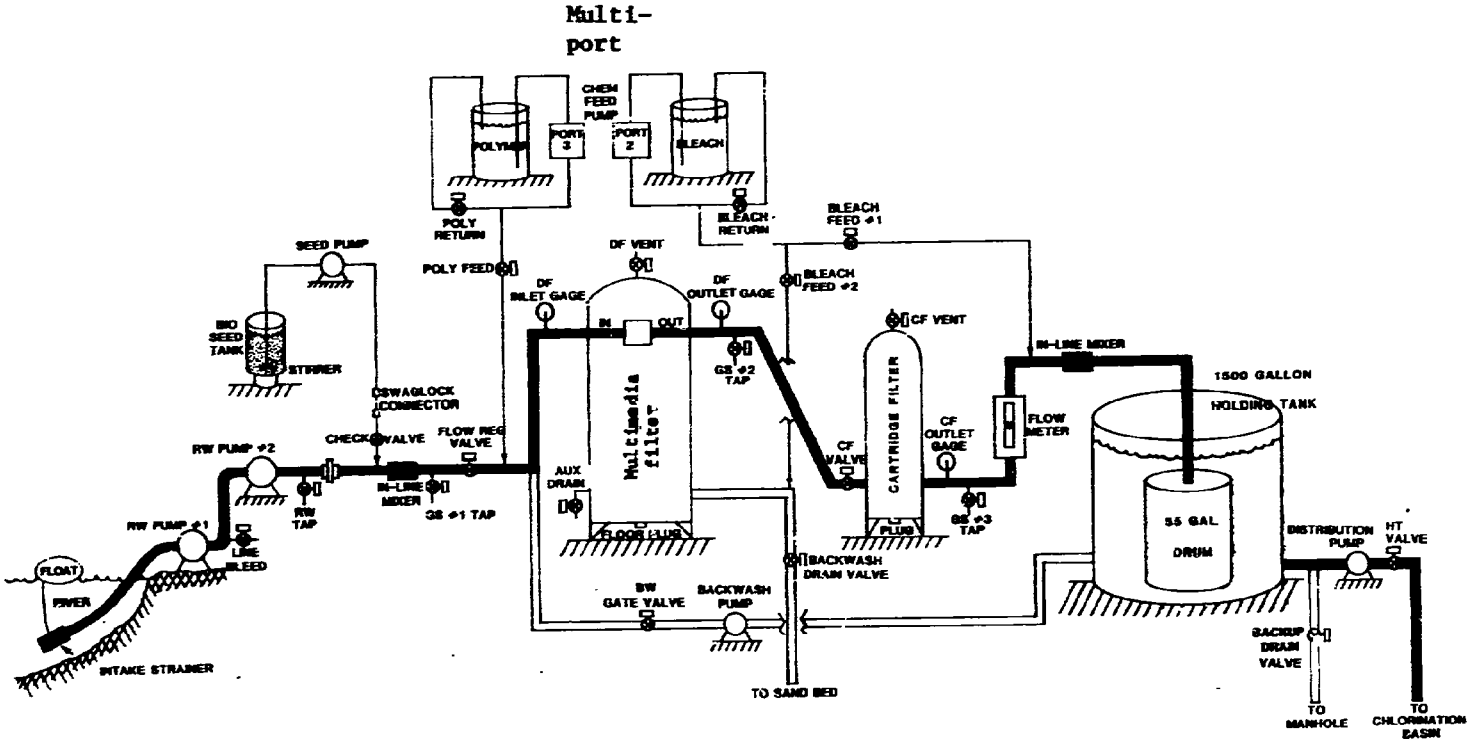


Figure 2. Experimental test layout.

TABLE 2. MICROORGANISMS USED IN FILTER EVALUATION

Organism[a]	Source
Bacillus globigii (BG)	Seeded
Escherichia coli (EC)	River
Poliovirus I (PV)	Seeded
Total Count	River
Total Enterics	River

a. See "Microbiological Indicators"

The standard multimedia filter (MMF) used in the ROWPU treatment system is a Culligan Model MD30 Mixed Media Filter. The filter uses four filtration media and two support media (Figure 3). The top filtering medium consisted of 3 inches of 1/8 x 1/8 inch plastic pellets having a specific gravity of 1.2 and a density of 45 pounds per cubic foot. The second filtration layer was of anthracite coal, 14 inches deep, having an effective size of 0.8 mm, a specific gravity of 1.5, a uniformity coefficient of 2, and a density of 52 pounds per cubic foot. The third filtration layer was made of 7 inches of calcined aluminum silicate having an effective size of 0.42 mm, a uniformity coefficient of 1.74, and a specific gravity of 2.5. The final filtration layer consisted of 3 inches of garnet sand, having a specific gravity of 3.95, a density of 135 pounds per cubic foot, an effective size of 0.3 mm, and a uniformity coefficient of 1.2. Two layers supported these media; the upper layer size-rated as G-12 garnet gravel and the lower layer rated as medium garnet gravel (coarser than G-12). The filter has a 30-inch internal diameter and, at a nominal 30 gpm flow rate, was loaded at 6.5 gpm per square foot.[5]

Water treated by the multimedia filter received additional treatment (polishing) in a cartridge filter (CF). The cartridge filtration system consists of six Filterite elements (see Appendix A) housed in a single body. These filter elements are constructed of woven polypropylene and are rated at 5 microns (nominal).

Following filtration the treated water was chlorinated and sent to a holding tank prior to discharge to the Fort Detrick Sewage Treatment Plant. Other equipment utilized in the experimental apparatus is listed in Appendix B.

A nominal 9-hour time frame per experiment was decided upon after consideration of field operational procedures, support logistics, and personnel requirements. The field operational procedures were patterned after those in the Technical Manual[6] and are described in Appendix B.

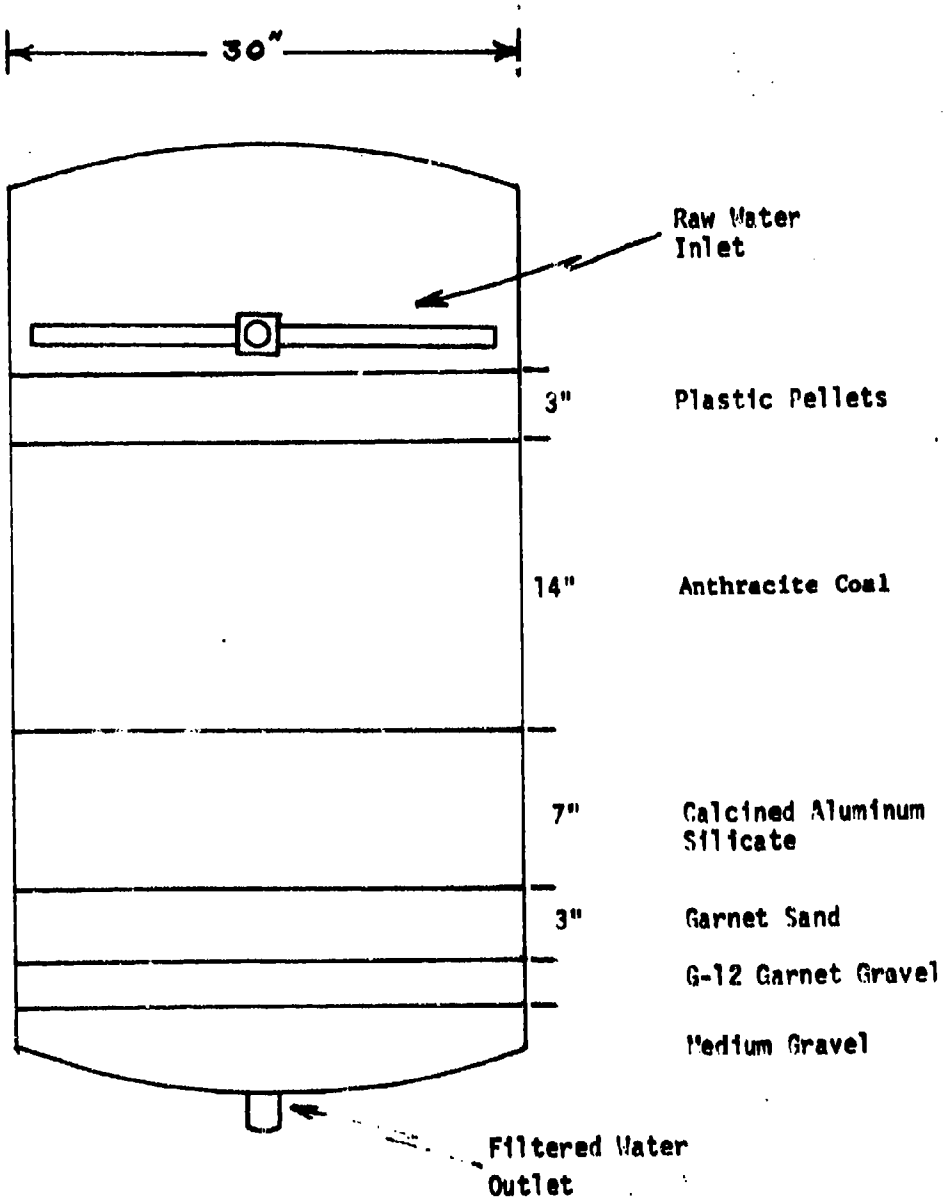


Figure 3. ROWPU multimedia filter.

EXPERIMENTAL DESIGN

The study was designed to conform statistically to a blocked multifactor test design.[7] The design is explained in detail in Appendix C. In such a design, experimentally imposed treatment factor combinations are applied once within a block of tests. One such factor was flow rate. Three rates were adopted: 30 gpm, the nominal 600 gph ROWPU intake flow rate; 35 gpm; and 40 gpm. The 40 gpm flow rate was determined to represent the maximum flow rate maintainable under the most extreme operating conditions.[6] Each day's test operated at one flow rate. During three closely-spaced tests, each flow rate was used once, and the trio of tests formed a block. Within each block, flow rate selection was random, a standard practice when uncontrolled external factors (e.g., turbidity) exist. Within each day's test, samples were drawn at fixed sample points (see Figure 2) as well as at fixed elapsed sampling times (t = 0, +3, +6, and +9 hours). This arrangement allowed for flexibility and efficiency in statistical design.

The statistical analyses proposed for the study were presented in the pre-test plan. A summer season, expected to involve river water of fairly low turbidity and consistently warm temperature, was scheduled for 1980. A spring season, expected to involve colder and more turbid river water conditions, was scheduled for 1981.

SAMPLING

Sample ports were provided in the test apparatus to collect water samples from the raw water feed line prior to microbiological seeding, in the raw water feed line following seeding, after multimedia filtration, and following cartridge filtration.

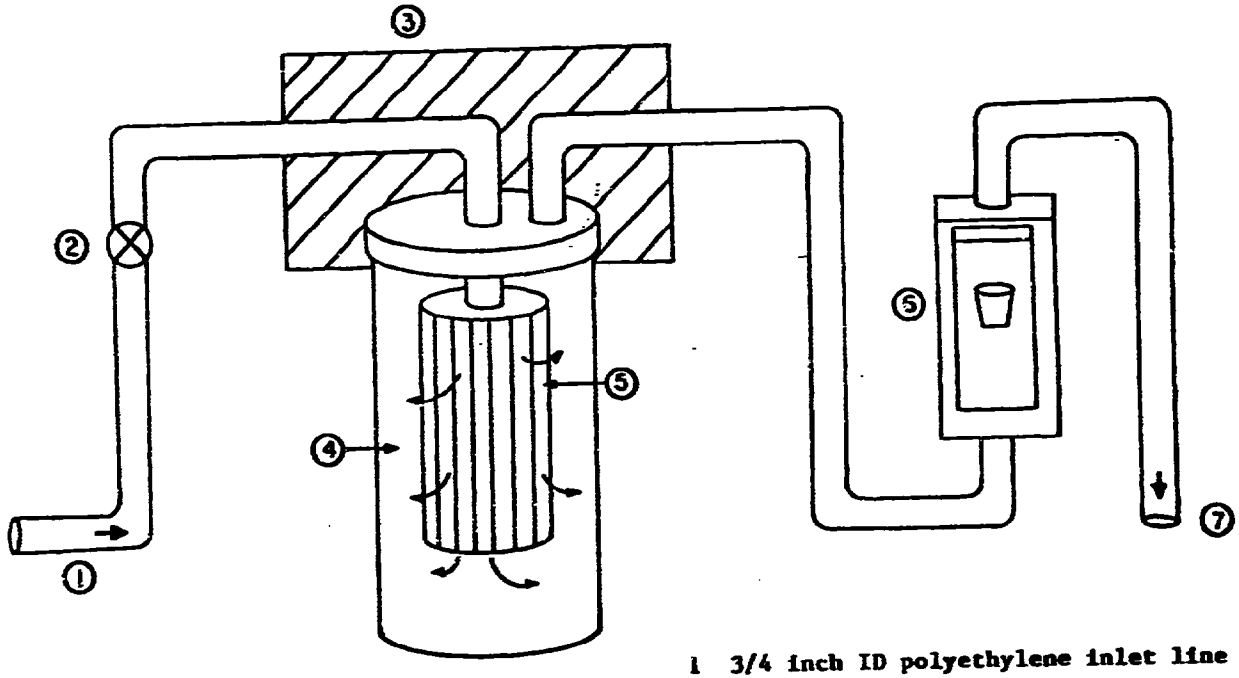
The sample schedule appears in Table 3. Four sampling periods were employed in each experiment: Start, +3 hours, +6 hours, and +9 hours of elapsed processing of a seeded water with acceptable product clarity. Each sampling followed the same order: seed tap, raw water tap, GS1 tap, GS2 tap, and GS3 tap (see Figure 2). With the exception of virus concentrate samples, grab sampling was used. A 10-minute delay was observed between GS1 tap and GS2 tap samplings to allow transient changes to be completed. Determination of this delay time is detailed in Appendix D.

Poliovirus (PV) was concentrated onto a Zeta Plus Cuno filter (see Appendix A) from 200-liter samples drawn directly from either the GS1, GS2, or GS3 tap. Line pressure was sufficient to maintain about 6 liter/min flow through the concentration apparatus. Collection time per sample was 30 to 35 minutes. The virus concentration apparatus used is diagrammed in Figure 4.

TABLE 3. SAMPLING SCHEDULE, INCLUDING SAMPLE VOLUMES

Parameter	Seed Tank	RW Tap	GS1 Tap	GS2 Tap	GS3 Tap	Sample Volumes
pH	N/D	N/D	All periods	N/D	All periods	As needed
TDS	N/D	N/D	All periods	N/D	All periods	As needed
Chlorine Demand	See App. B. Sect. III 3.	N/D	All periods	N/D	All periodsd	As needed
TOC	N/D	N/D	All periods	N/D	All periods	250 mL
Turbidity	N/D	N/D	All periods	All periods	All periods	250 mL
Alkalinity	N/D	N/D	All periods	N/D	All periods	Use turbidity sample
E. coli	All periods	0,9	All periods	All periods	All periods	b,c
B. globigii	All periods	N/D	All periods	All periods	All periods	Use *E. coli* sample
Total counts	N/D	0,9	All periods	All periods	All periods	Use *E. coli* sample
Total enterics	N/D	0,9	All periods	All periods	All periods	Use *E. coli* sample
PV concentrate	N/D	N/D	6	3,9	3,9	200 L
PV direct	All periods	N/D	All periods	3,9[d]	3,9[d]	9 mL

a. N/D = not done.
b. Ten-milliliter sample from seed tank.
c. One-liter sample from nonseed tank collection points.
d. Sampled during Block 2 tests only.



1 3/4 inch ID polyethylene inlet line
2 Flow regulation valve
3 Flow manifold (in section)
4 Filter housing
5 Filter cartridge
6 Flowmeter
7 Outlet line to collection tank

Figure 4. Virus concentration apparatus.

The following scheduling procedure was followed.

- When PV concentrate was collected from the GS1 tap, collection immediately followed all other sampling at this tap.
- When PV concentrate was collected from the GS2 or GS3 taps, collection started immediately preceding the sampling at the GS1 tap. The GS2 and GS3 taps had two outlets each so that independent grab samples could also be collected.
- The +9 hours samples actually occurred at 8 1/2 to 8 3/4 hours after the start samples at each tap to allow time for the PV concentrate collection at the GS2 and GS3 taps.

PHYSICAL AND CHEMICAL ANALYSIS

The physical and chemical analyses used in this study are summarized in Table 4. Total dissolved solids, pH, and chlorine demand were determined on site during the testing. Other analyses were conducted at the fixed laboratories of USAMBRDL.

TABLE 4. PHYSICAL/CHEMICAL ASSAY SUMMARY

Assay	Location	Method	Units	Remarks
pH	On site	Ion-electrode	--	
Total Dissolved Solids (TDS)	On site	Conductivity meter with TDS scale[a]	ppm	See Appendix F
Chlorine demand	On site	FACTS test analysis of sample titrated with NaOCl solution	ppm	See Appendix F
Alkalinity	Lab	Standard Methods 403,[8] pH 3.7 end point	mg/L $CaCO_3$	
Turbidity	Lab	Standard Methods 214A[8]	NTU	
Total Organic Carbon (TOC)	Lab	Standard Methods 505[8]	mg/L	Sample acidified by H_2SO_4 in field, see Appendix F

a. Model DP-03, Devon Products Corporation, Los Angeles, CA.

MICROBIOLOGICAL ANALYSIS

Microbiological Indicators

Three species of microorganisms were selected for specific enumeration: Bacillus globigii (BG), Escherichia coli (EC), and the LSc strain of Poliovirus I (PV). Bacillus globigii spores and PV were added to raw river water from the Bio Seed Tank (see Fig. 2). Preliminary river tests indicated that the background EC levels in the river were sufficiently high to permit use of the river as the EC source.

Bacillus globigii was available as a dry spore suspension, 800 grams of material with a titer of approximately 10^{11} colony forming units per milliliter (CFU/mL). The organism is a simulant of Bacillus anthracis. In nature, BG is a soil organism and occasionally occurs in water due to storm runoff. The assay method followed the dilution and spread plate technique.[9]

Escherichia coli is a common enteric bacterium, used as a standard indicator of water contamination. The dilution spread plate method was used to assay samples for EC on eosin methylene blue agar.[9]

The LSc strain of Poliovirus I is used for oral polio vaccine. Poliovirus has been isolated in human fecal discharges and can survive the sewage treatment process to enter surface waters.[10] For these reasons, PV was chosen over more easily assayed indicator viruses, such as the f2 bacteriophage. Enumeration of PV was by infection of a monolayer of HeLa cells followed by agar overlay.[11]

Two indigenous bacterial populations were assayed as part of the study: standard plate count (total count) and total enteric bacteria (total enterics). The total count enumerates bacteria capable of aerobic growth on agar. Because this was the same assay method as used for BG, no new procedures were needed. The total enteric count involves those organisms present in a water source that grow on eosin methylene blue agar at 35°C. Such organisms are found in fecal waste discharges, although they are not necessarily human in origin.

Microbiological Growth Procedures and Preparation

The design goals for BG and PV content in seeded raw water had to be compatible with supplies on hand, production capabilities, and sampling volume-processing capabilities in the field. These goals were set at 10^4 CFU/mL for BG and 10^2 plaque-forming units per mL (PFU/mL) for PV. At such levels, even four log removal (99.99%) by the prefiltration system could be observed by sampling procedures that were manageable. Based on a 9-hour test (the test time adopted) and a 40 gpm maximum flow, approximately 8×10^{11} CFU of BG and 8×10^9 PFU of PV were required per test. For each test, 1.5 grams of dried BG spores were dispersed in 500 mL of phosphate buffered saline solution to form the spore suspension. The solution was then transferred to the water in the Bio Seed Tank.

Poliovirus I had to be cultured on live mammalian cells. The HeLa cell strain was selected and grown to confluence in a 850 cm^2 (surface area) roller bottle. The cells were inoculated with PV from seed stock with a multiplicity of infection of 10:1. The cells were exposed to virus for 1 hour; then fresh medium was added on the cells. The roller bottles were incubated at 35°C until cell lysis indicated virus production (usually 18 to 24 hours). The virus was harvested by centrifugation, the supernatant containing the virus. Products from several roller bottles were composited, an aliquot was saved for titer determination, and the remainder was frozen at -75°C until required for use. For each test, a frozen composite was defrosted in a 35°C water bath. On the basis of the aliquot titer, a suspension of sufficient volume to provide 8×10^9 PFU was added to the water in the Bio Seed Tank.

Sample and Assay Procedures

Samples collected for bacterial assay were either 10 mL or 1 liter. For the 10-mL samples, 30 mL screw-cap test tubes were used; for the 1-liter sample, a 1-liter polypropylene bottle was used. Both tubes and bottles were sterilized and kept closed until sample collection time. Samples collected for viral assay were either 9 mL direct samples or concentrated from 200 liters. For the 9-mL sample, 1 mL of sterile 10X Hank's Balanced Salt Solution was added. Each 200-liter sample was processed through a sterile Zeta Plus Cuno Filter. Preliminary studies at USAMBRDL indicated that the concentration efficiency of the filter for poliovirus was 94 percent.

After the concentration procedure was completed, the Cuno Filter was immersed in 600 mL of sterile 3 percent beef extract solution at pH 9.5. This treatment eluted the virus from the filter cartridge. The eluate was adjusted to pH 3.5, forming a floc. The floc was stirred for 15 minutes and centrifuged at 10,000 x g for 20 minutes. The supernatant was discarded. The pelleted virus was resuspended in 10 mL of 0.15 M $Na_2HPO \cdot 7H_2O$ solution and frozen until assay. The unconcentrated samples were also frozen until assay.

For the assay of PV, HeLa cell monolayers were established on 60 x 15 mm tissue culture plates. Virus samples were diluted using 1 mL of sample to 9 mL of incomplete medium blanks containing Minimal Essential Media (MEM) and Hank's Balanced Salt Solution. Serial dilutions were prepared in a similar manner. The serially diluted samples of 0.2 mL volume were pipetted on monolayered HeLa cultures, with rocking every 15 minutes to assure infection. After exposure for 1 hour, the infected plates were overlaid with 1 percent agar containing MEM and incubated at 35°C in an incubator for 3 days prior to plaque count. The plates were then stained with neutral red to enhance plaques for counting. A 95 percent CO_2:5 percent air atmosphere was maintained in the incubator.

Bacterial assays were accomplished by the dilution and spread plate method. Vortex mixing was used to assure uniform suspensions in dilution tubes. Petri dishes were prepared, each containing 10 to 15 mL of the specified agar. For the assay of total counts and BG, Standard Methods Agar was used; for EC and total enterics, Eosin Methylene Blue Agar was used. To each plate, 0.1 mL of sample (or serial dilution) was added and spread. The petri dishes were incubated for 18 to 24 hours and counted. Bacillus globigii was enumerated in the presence of other bacteria of the total count group by its colony morphology and distinctive orange coloration. Escherichia coli was

enumerated in the presence of other bacteria of the total enteric group by its non-mucoid green-sheen colored colonies.

RESULTS AND DISCUSSION

Equipment shakedown started August 1980, and testing commenced in mid-September. After three replicated blocks of testing, analysis of data collected by that time indicated that spring testing was not essential to achieve the objectives of this study. Appendix C includes a discussion of the statistical implications of this truncation of effort.

OPERATIONAL DATA

Temperature and precipitation trends for the first half of 1980 were approximately normal. The summer season turned out to be one of the warmest on record, and in August rainfall fell to about half the normal monthly level. By October, the trend of above-normal temperatures ended, but rainfall continued below normal. The consistent low-flow situation meant that the river exhibited relatively low turbidity. This was disappointing from an operational viewpoint but was beneficial from a statistical viewpoint because of minimal day-to-day variability due to varying river flow.

Table 5 summarizes the operational data for the three blocks. The flow rates, water temperatures, and pressure readings were recorded hourly during the course of each day's test. The polymer dosage was calculated based on the polymer content in solution and the field-calibrated flow rate (see Appendix A). Technical Manual instructions for an initial setting correspond to a 5.2 mg/L dosage.

The pressure drops observed across the multimedia filter on most test days did not reflect any trend indicative of heavy suspended solids loading. From an operational viewpoint, the multimedia filter was not subjected to a rigorous test throughout its performance range. A similar conclusion applied to the cartridge filter. While the multimedia filter was backwashed after each day's test, the cartridge filters were not. If appreciable suspended solids had been encountered by the cartridge filter, the pressure drop across the filter would have increased throughout the course of the study. This was not observed.

PHYSICAL AND CHEMICAL DATA ANALYSIS

The physical and chemical assay data of the three test blocks appear in Appendix F. Turbidity data were analyzed by the analysis of variance (ANOVA) technique (see Appendix C) in terms of the statistic log (turbidity). The effects noted with p-value <0.05 were elapsed time in a test, sample point, and the interaction of these effects. No flow effect was statistically detectable. These effects are noted by the four trend patterns of Figure 5. Specifically, the $t = 0$ turbidity reduction was lowest, and as the test progressed, product water became clearer. The percent reductions in turbidity, with 95 percent confidence limits of the mean percentage reductions, are listed in Table 6. The interaction effect is noted by comparing the four trend patterns of Figure 5. The cartridge filter removal was marginal relative to that achieved with the multimedia filter. The bulk of the turbidity removal occurred across the multimedia filter.

TABLE 5. OPERATIONAL DATA

Parameters	Block 1			Block 2			Block 3		
	9/17	9/18	9/23	10/9	10/10	10/15	10/22	10/23	10/29
Nominal flow rate (gpm)	30	35	40	40	30	35	35	40	30
Flow rate range (gpm)	30	34.5–36	40–40.5	40–41	30–30.5	35–35.5	34.5–35.5	39.5–41	30–30.5
Air temperature (°C)									
Low–	16–	16–	22–	13–	8–	8–	9–	2–	2–
High	30	30	32	26	25	20	22	18	12
Water temperature (°C)	19.4–22.3	20.6–22.8	22.2–24.0	14.4–17.7	14.2–14.8	11.2–13.8	12.7–14.8	10.6–12.5	10.0–12.4
Polymer dosage (mg/L)	4.28	5.19	5.14	5.02	5.59	4.32	4.78	5.16	4.70
Pressures (psi)									
MMF-In[a]	13–13.5	16.5	18.5–20.5	19–20.5	12.5–13	16.5–17	16.5–17.5	20–21.5	13–14
MMF-Out	9–9.5	11–12	14.5–15	14.5–15.5	8.5–9	11.5–12	11.5–12.5	14.5–15.5	9–9.5
CF-Out[b]	8.5–9	11–12	14–15	14–15	8.5–9	11.5–12	11–11.5	14–15	8.5–9
MMF range	4–4.5	4.5–5.5	4–5.5	4.5–5	4	5	4.5–5	5–6	4–4.5
CF range	0–1	0–0.5	0–0.5	0–0.5	0	0–0.5	0–1	0.5–1	0.5–1

a. MMF = multimedia filter.
b. CF = cartridge filter.

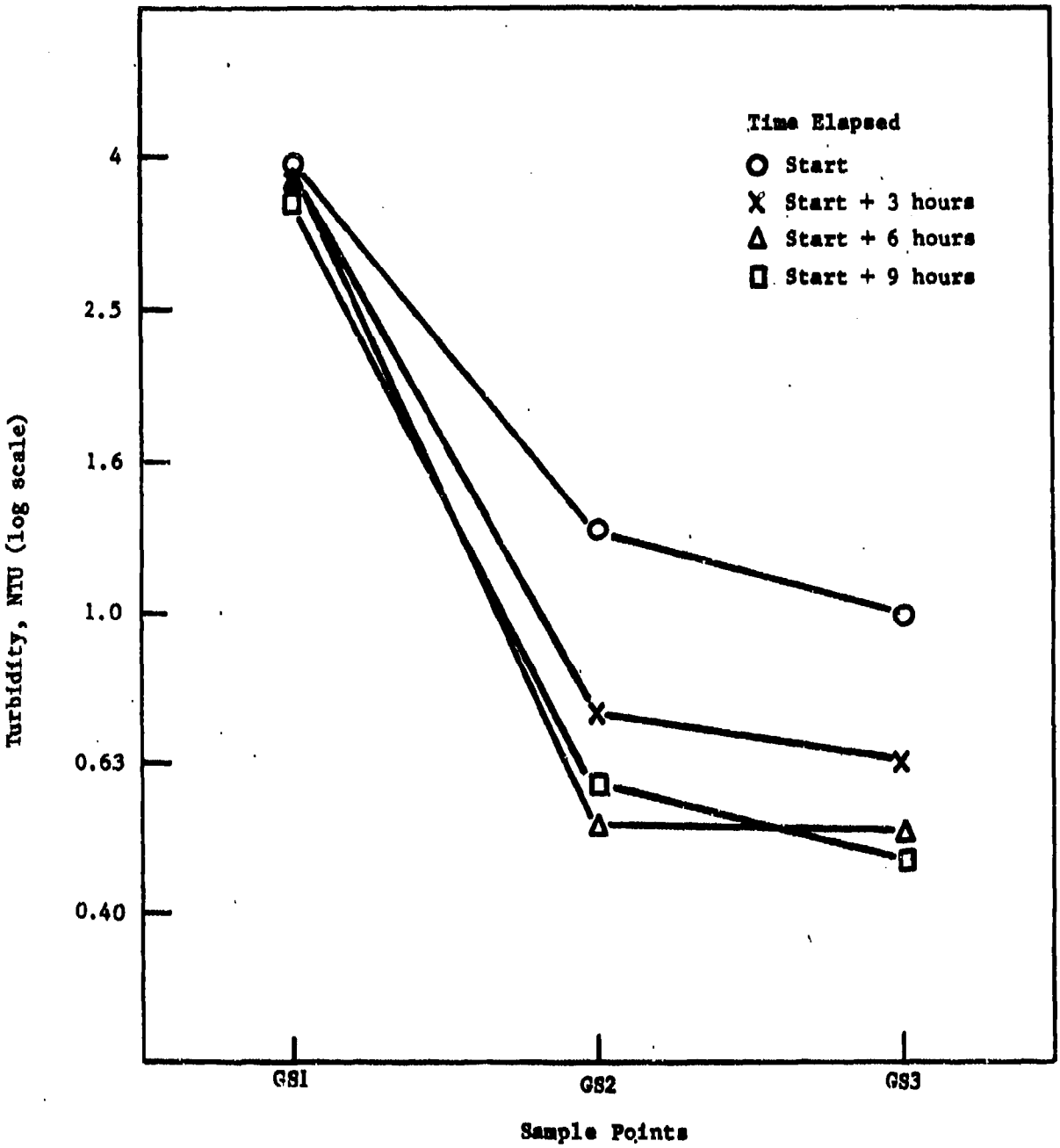


Figure 5. Graphical representation of the turbidity statistical analysis showing time effects.

TABLE 6. PERCENT REDUCTION IN LOG TURBIDITY FOR THE ROWPU PREFILTRATION SYSTEM

Elasped Time (hr)	Across Multimedia Filter		Across System	
	Mean	95% Confidence Limit	Mean	95% Confidence Limit
0	67.3	55.3 - 76.2	74.4	62.7 - 82.5
3	81.4	74.7 - 86.3	84.0	77.4 - 88.7
6	83.5	78.6 - 87.3	86.4	82.1 - 89.7
9	85.9	82.0 - 89.0	88.1	84.5 - 90.8

Chlorine demand was reduced from a mean concentration of 1.42 mg/L at GS1 to 0.83 mg/L at GS3. The 0.59 mg/L removal had an associated p-value of 0.052, which in view of the filter's suspended material removal, probably indicates a real trend. No statistically significant flow- or time-related effects were found. The 95 percent confidence limits on the mean removal were 0.36 and 0.82 mg/L.

A mean TOC removal of 1.4 mg/L was computed. This was based on a mean 5.1 mg/L TOC in system influent (4.6 mg/L in raw water and 0.5 mg/L in polymer) to 3.8 mg/L at GS3. This removal can only be considered a rough estimate in view of the precision of the assay data.

The other physical parameters were not analyzed for system influence. There was a daytime increase in pH during most test days. Alkalinity tended to increase during the study, probably as a consequence of cooling river water. Total dissolved solids levels were well below the unacceptable 1,500 ppm level.[6] Alkalinity and TDS levels on October 29 were probably a consequence of run-off from a heavy rain on October 25.

MICROBIOLOGICAL DATA ANALYSIS

The prefiltration system's microbiological removal performance was based on an analysis of variance (ANOVA) of log (assay) data along with an evaluation of specific assays. The assay data for BG, EC, PV, total counts, and total enterics appear in Appendix F, Tables F-3 to F-7, respectively. The PV assays subjected to ANOVA were those of the concentrated samples. The details of the ANOVA are in Appendix C1. In the ANOVA, the log (assay) observations are assumed to be log-normally distributed. The summary ANOVA results, expressed in terms of p-values for hypothesized treatment effects, are in Table 7. An alpha level of 0.05 was selected for investigating the statistical significance of effects, a level commonly employed in ANOVAs.

TABLE 7. ANALYSIS OF VARIANCE SUMMARY: p-VALUE OF EFFECTS

Effect	p-Value for Indicated Microorganism/Group				
	BG	EC	PV	Total Count	Total Enterics[a]
Sample Point (S)	0.0001	0.0001	0.0012	0.0001	0.0001
Paired Comparisons					
GS1-GS2	0.0001	0.0001	0.0005	0.0001	0.0002
GS1-GS3	0.0001	0.0001	0.0029	0.0002	0.0001
GS2-GS3	0.262	0.309	0.328	0.222	0.620
Flow Rate (F)	0.052	0.877	0.980	0.035	0.639
Elapsed Time (T)	0.224	0.086	[b]	0.066	0.147
Interactions					
FxS	0.221	0.659	0.449	0.004	0.413
SxT	0.556	0.116	[b]	0.816	0.365
FxT	0.414	0.173	[b]	0.354	0.519
FxSxT	0.563	0.199	[b]	0.848	0.801

a. Based on Block 2 and Block 3 assays only (see Appendix C-3).
b. Sampling plan not applicable to analysis of these effects (see Appendix C-3).

There is a distinction between sample point (S), elapsed time (T), and flow (F) effects. A statistically significant sample point effect relates to system performance; there is a statistically significant difference between the log (means) of two or more sample points. The analysis used allows three paired comparisons to be assessed: GS1-GS2 (the MMF), GS2-GS3 (the CF), and GS1-GS3 (the prefiltration system). If statistically significant p-values are noted for any of these comparisons, there is statistical evidence of a difference between the log (means) of the cited sample points. Elapsed time and flow effects are estimated from pooled sample point values and do not translate readily into operational parameters. If flow or elapsed time effects are significant, they pertain to system performance when their interactions with sample point (FxS, SxT, FxSxT) are statistically significant. Otherwise, the cause of such effects may be more likely to be external to system performance and may not be necessarily of operational importance.

Bacillus globigii Analysis and Discussion

The raw data (Table F-3) indicate variability from run to run in the Bio Seed Tank concentration. This in turn directly affects the number of BG CFU/mL assayed at GS1. The variability is inherent in assaying BG, because of the process involved in the manufacture of BG spores. The organism is grown in liquid medium, which is spray dried, resulting in microscopic-sized aggregates with varying number of spores heterogeneously mixed with dried medium. Although the amount of medium weighed from run to run was 1.5 grams,

the number of BG spores was not consistent. The multifactor analysis statistically removes the effect of such variation from paired comparisons.

The raw data also indicate variability in the trend of BG CFU/mL reduction across sample points for each specific sample set. A reduction was anticipated across each filter. A decrease in BG CFU/mL consistently occurred across the MMF. However, there were several instances where an increase in BG CFU/mL occurred across the CF. A possible explanation for this is that such samples were taken during a period of breakthrough. That is, as bacterial particles filled the CF interstitial spaces, the local fluid velocity increased until the particles were dislodged and passed through the filter.

The BG ANOVA is in Table 7. As indicated, the reduction in log means across the MMF (GS1 vs. GS2) and across the prefiltration system (GS1 vs. GS3) was determined to be statistically significant ($p < 0.05$). However, the reduction across the CF (GS2 vs. GS3) was not determined to be statistically significant. This means that the reduction in BG is due primarily to the MMF.

A marginal effect due to differences in overall flow rate means was detected ($p = 0.052$). However, the absence of a FxS interaction indicates no strong statistical evidence of differences between flow rate reductions across the sample points (Table C-2 and Fig. 6). No elapsed time or SxT interaction effects were noted. A graphical display (Fig. 6) of the flow rate profile composited over time indicates an upturn at the 40 gpm flow rate from GS2 to GS3. However, the trend was not strong enough relative to the standard error of the means to be detected statistically as a FxS interaction.

The overall log reductions in BG CFU/mL appear in Table C-2 and are displayed graphically in Figure 7. Percent reductions are presented in Table 8 along with 95 percent confidence intervals. These intervals indicate that the statistical variability of the BG reductions was the least of all microorganisms or groups analyzed. Percent removals approached 99 percent for BG.

Escherichia coli Analysis and Discussion

Contrasted to BG and PV, the EC challenge to the system was from indigenous organisms in river water. Accordingly, the range in GS1 assays reflects uncontrollable fluctuations in river EC content. Over the entire test period, GS1 assays of 24 to 8,500 CFU/mL were noted. Even within a given test day, changes as high as 28-fold (compare 255 vs. 7,280 on Sep 23) were observed. One interesting observation was that t = 0 assays were usually higher than at other times; in great part this is reflected in the elapsed time effect p-value in Table 7.

As with BG, there were several specific times when CF effluent had much higher EC content than CF influent.*

* These assays are, with means (back-transformed from log-means in Table C-3):

Date	Time	GS1	GS2	GS3
Sep 23	t = 0	7,280	165	1,180
Sep 23	t = 3	3,740	24	346
Mean		189	17	12

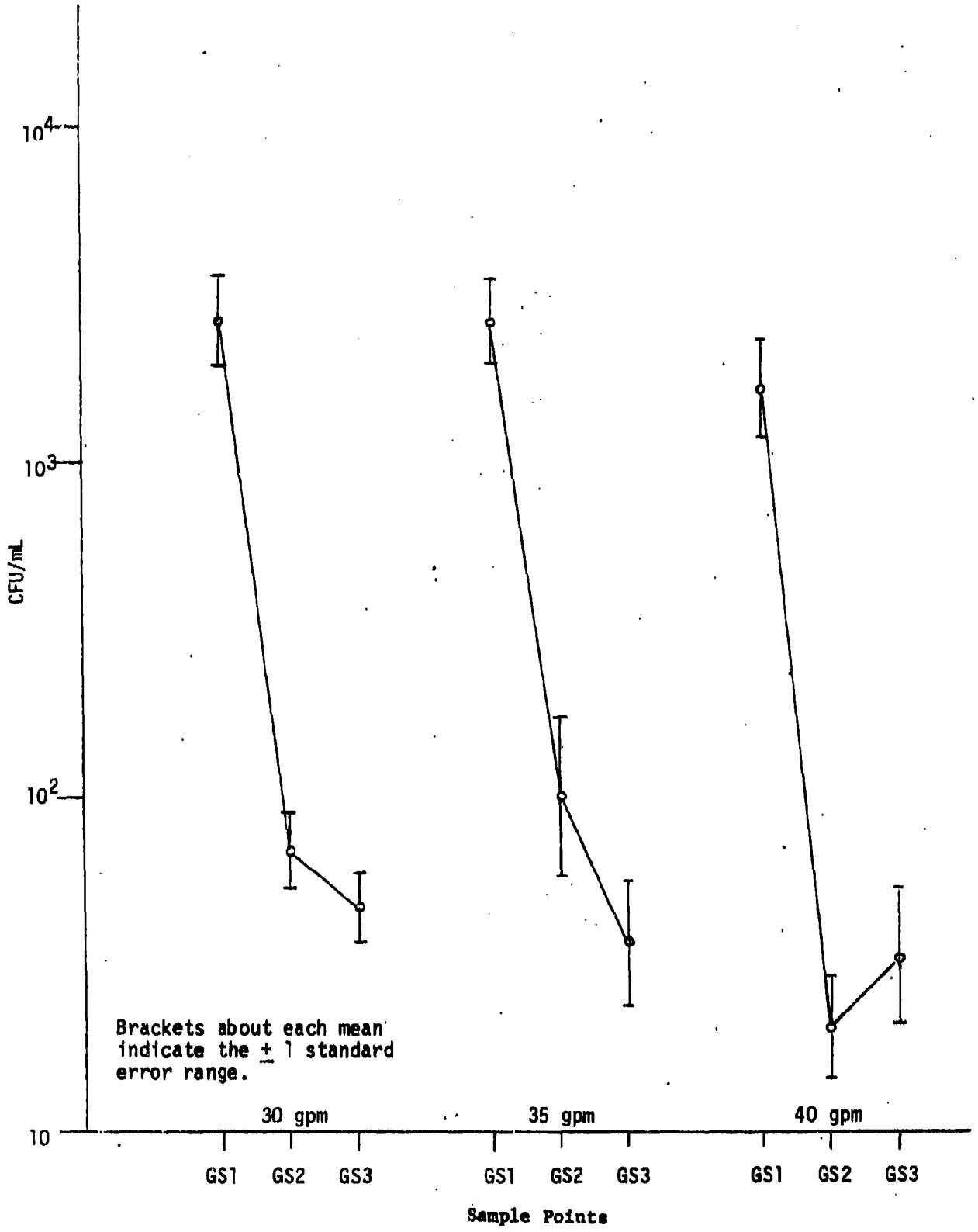


Figure 6. Mean Bacillus globigii reduction through prefiltration system by flow rate.

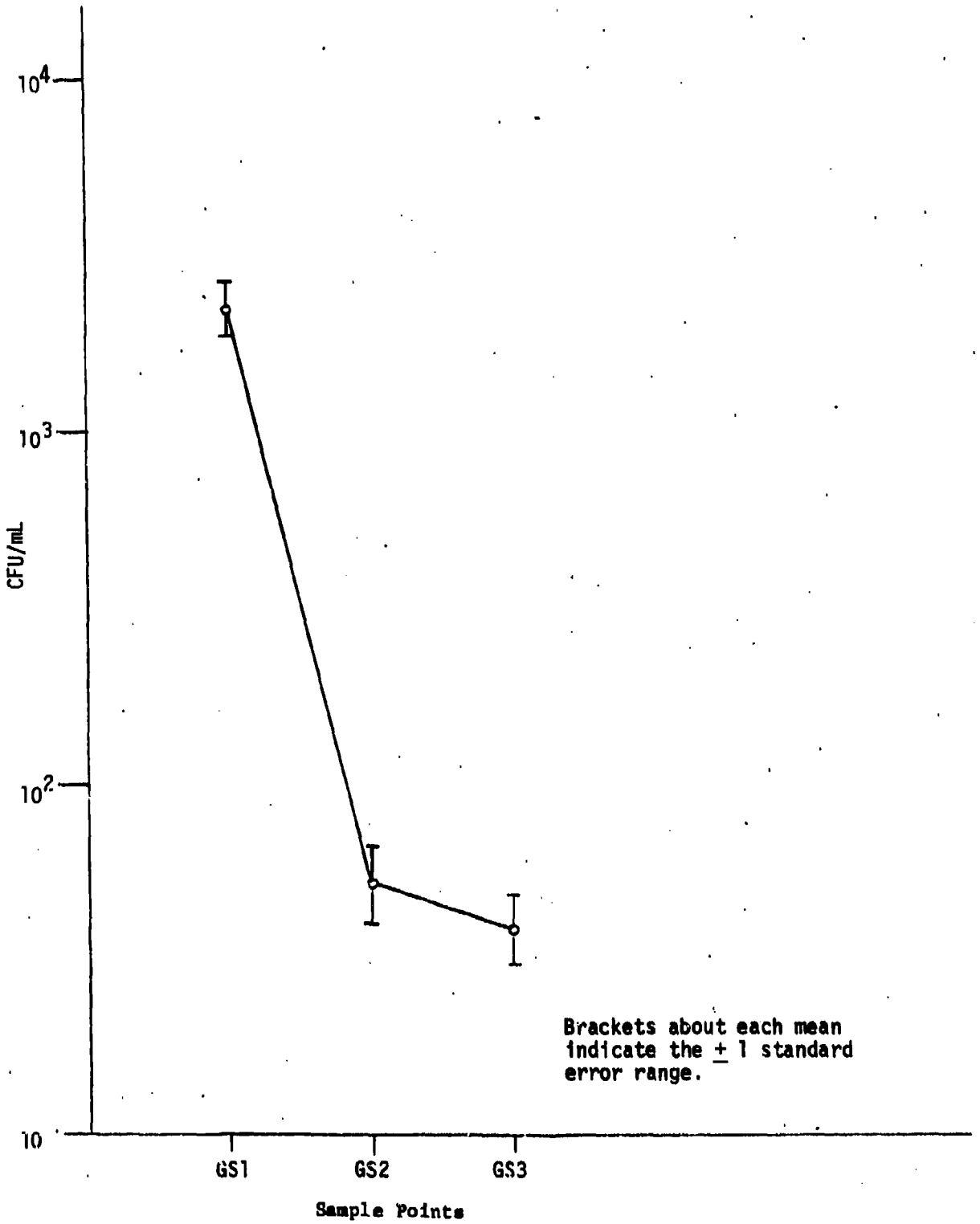


Figure 7. Summary graph: Mean Bacillus globigii reduction (flow rates pooled) through prefiltration system.

As with BG, these events occurred during 40 gpm flow rate operations; thus, the breakthrough discussed with BG may also apply to EC.

The ANOVA for EC (Table 7) indicates that only sample point effects have a p-value <0.05, and that from the paired comparisons, the MMF is the prime unit effecting EC removal. The log-mean reductions of EC at the sample points are shown in Figure 8; these log-means, partitioned for flow, are presented in Figure 9. Data for both figures are in Table C-3. Percent removals and 95 percent confidence limits calculated from these data are in Table 8. System performance is more erratic than that estimated for BG; the across-system 95 percent confidence limits represent a range from 0.86 to 1.44 logs of removal.

TABLE 8. SUMMARY OF PERCENT REMOVALS

Microorganism/ Group	Across MMF		Across CF[a]		Across System	
	Mean %	95% Confidence	Mean %	95% Confidence	Mean %	95% Confidence
BG	97.7	96.1 98.6	0.6	-1.5 2.9	98.3	97.1 99.0
EC	91.0	83.2 95.1	2.5	-7.7 13.2	93.5	87.4 96.4
PV	72.0	44.6 85.9	8.3	-25.0 45.9	80.3	60.9 90.5
Total Count	74.4	52.5 86.2	8.7	-23.3 39.8	83.1	62.9 92.3
Total Count 30 gpm	86.2	56.4 95.6	3.3	-28.8 40.3	89.5	66.8 96.7
Total Count 35 gpm	44.6	-75.3 82.5	48.4	-4.5 >100[b]	93.0	78.0 97.8
Total Count 40 gpm	78.1	30.5 93.1	-44.9	-204.7 48.4	33.2	-111.6 78.9
Total Enterics	83.7	62.5 93.0	3.1	-23.4 21.8	86.8	69.6 94.3

a. Mean % expressed in terms of system influent count. The across CF 95% confidence limits are roughly approximated in terms of the 95% confidence performance across MMF and across system. For BG, these are: upper limit, 99.0%-96.1% = 2.9%; lower limit, 97.1%-98.6% = -1.5%.

b. More CFU/mL are expected to be removed across the CF than were in system influent.

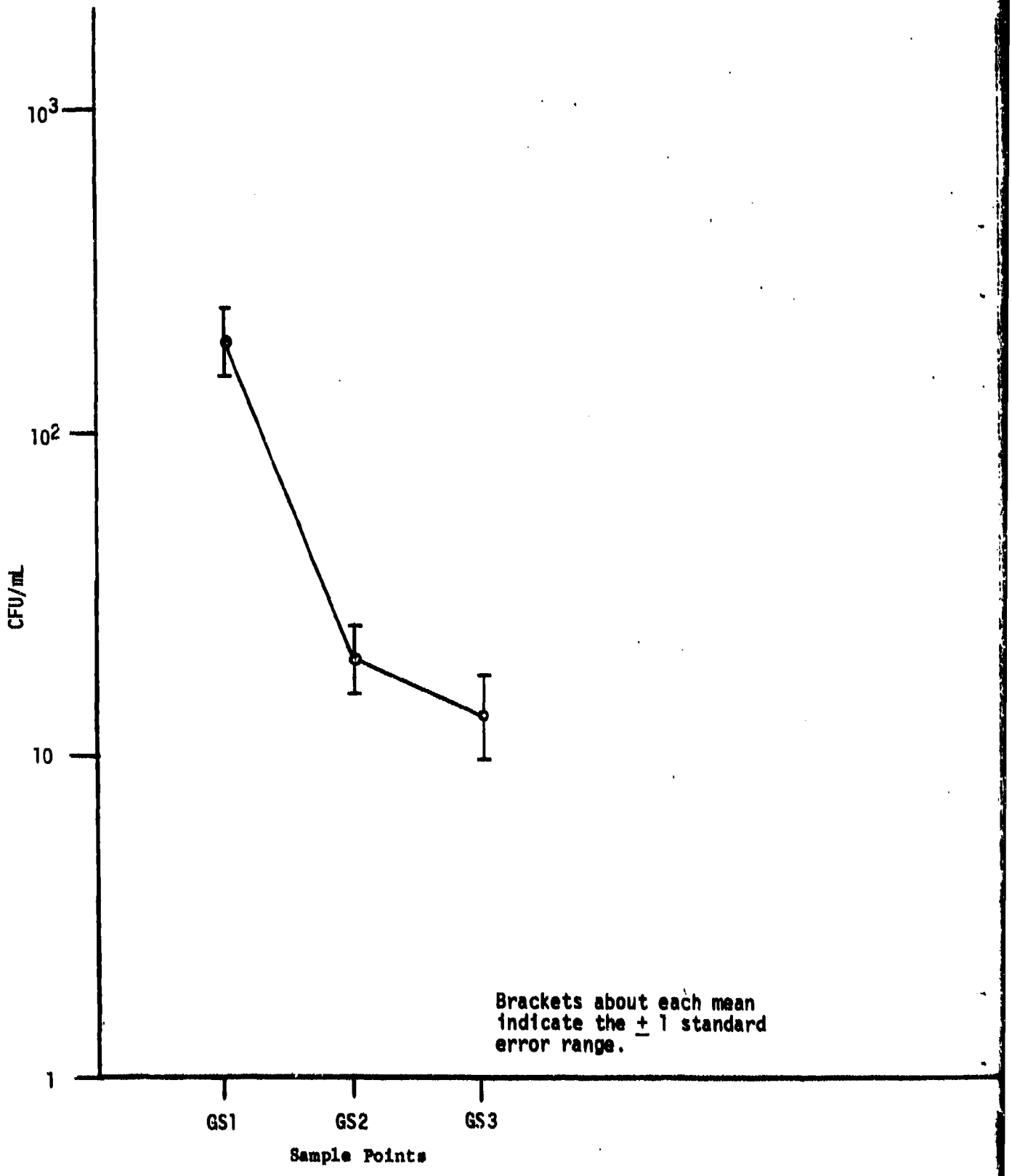


Figure 8. Summary graph: Mean Escherichia colli reduction (flow rates pooled) through prefiltration system.

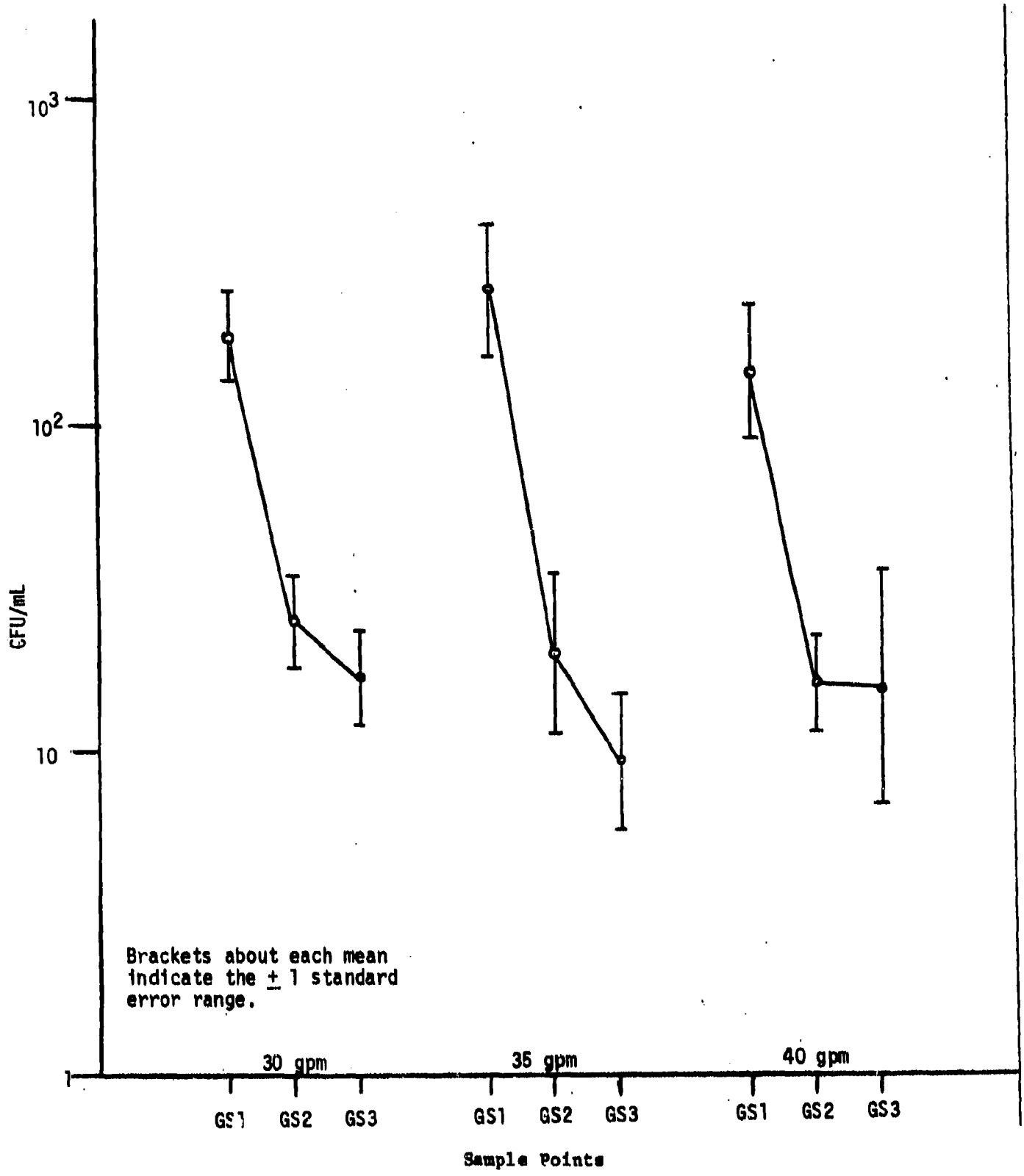


Figure 9. Mean Escherichia coli reduction through prefiltration system by flow rate.

Poliovirus: Analysis and Discussion

As shown in Figure 10, the flow-pooled mean poliovirus removal computed can be mainly attributed to the MMF. Removal across the system was 80.3 percent (Table 8). Figure 11 indicates PV removal at each flow rate. Although not statistically significant, there is a graphical indication at 40 gpm that the CF was possibly allowing breakthrough.

The flow and flow X sample were not factors in the reduction of PV across the system as statistically analyzed. The p-value for flow was 0.980 and the p-value for flow X sample was 0.449. As to the sample locations (GS1, GS2, and GS3), the major portion of the reduction of PV occurred through the MMF ($p = 0.0029$). A statistically insignificant amount of reduction occurred due to the cartridge filter ($p = 0.328$).

In 1974, Ford and Pressman[4] studied the passage of bacteriophage f2 through the prototype ROWPU, utilizing the accepted laboratory methodology at that time. Their results differ from the present study, as they reported high removal rates; see Table 1. However, removal rate comparisons are of dubious validity. Ford and Pressman utilized a bacterial virus; this study utilized a mammalian virus. The isoelectric points of these viruses differ; they would be expected to respond differently to a coagulant (both studies used Catfloc-T). The methods of collection and assay differed. Ford and Pressman did not concentrate samples (the technology was not available in 1974). Also, their experiments were conducted on a dual-media prototype filter, not the ROWPU MMF. The major difference in technique was the appearance of virus concentration technology in the late 1970s. The virus concentration methodology lends itself to detection of virus at levels that were heretofore undetectable.

Microbiological Groups: Analysis and Discussion

The total count assay used detected BG and background aerobic organisms. Some of these organisms were also represented in the total enteric count. Moreover, the total enteric group includes EC. As contrasted to the specific microorganism assay, these groups are statistically dependent; that is, organisms from the total enteric assay influence the total count assay.

The total count ANOVA of Table 7 indicates a flow, sample point and FxS effect with p-values <0.05. The paired comparisons, which are pooled over time and flow, indicate that the MMF filter is the main unit causing removal. This is graphically illustrated in Figure 12; see Table C-5 for data base. The corresponding percent removals and 95 percent log mean confidence levels appear in Table 8. The strong FxS interaction suggests that removal performance was flow-related. This is graphically illustrated in Figure 13. Three relative observations could be made: MMF removal performance at 35 gpm was poor; CF removal performance at 35 gpm was good; CF removal performance at 40 gpm was poor. Accordingly, the percent removals in Table 8 are presented at each flow. The main flow effect does not relate to system performance; see Appendix C-3.

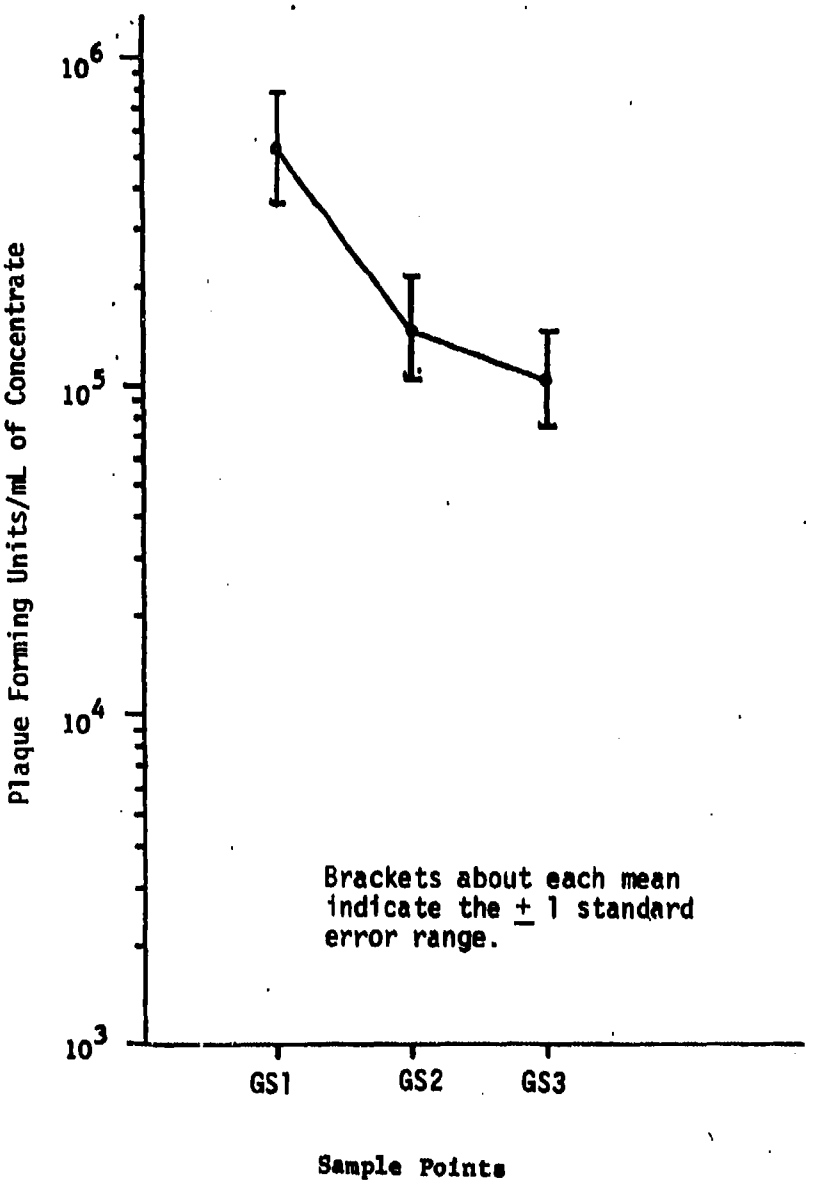


Figure 10. Summary graph: Mean poliovirus reduction (flow rates pooled) through prefiltration system.

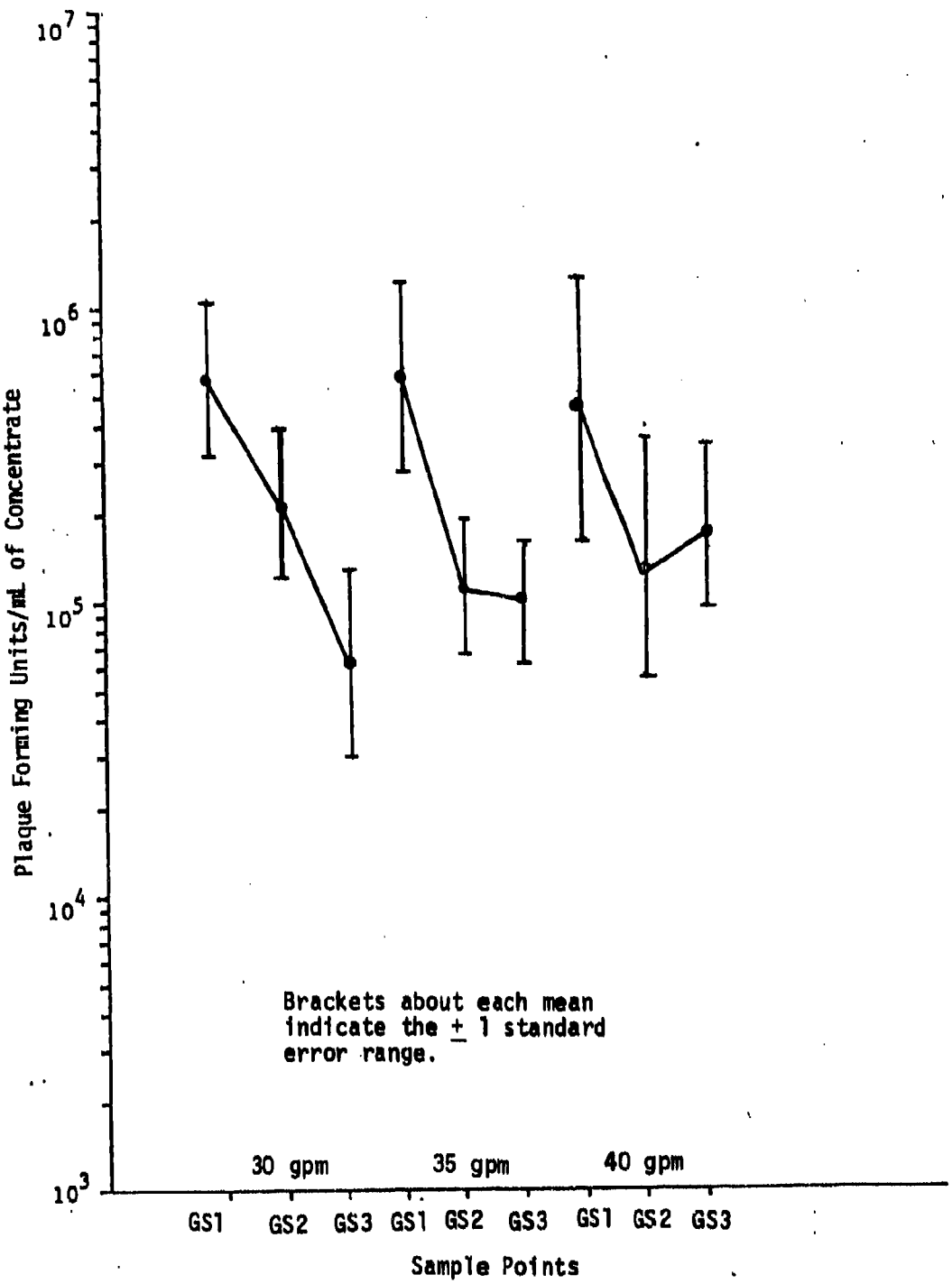


Figure 11. Mean poliovirus reduction through prefiltration system by flow rate.

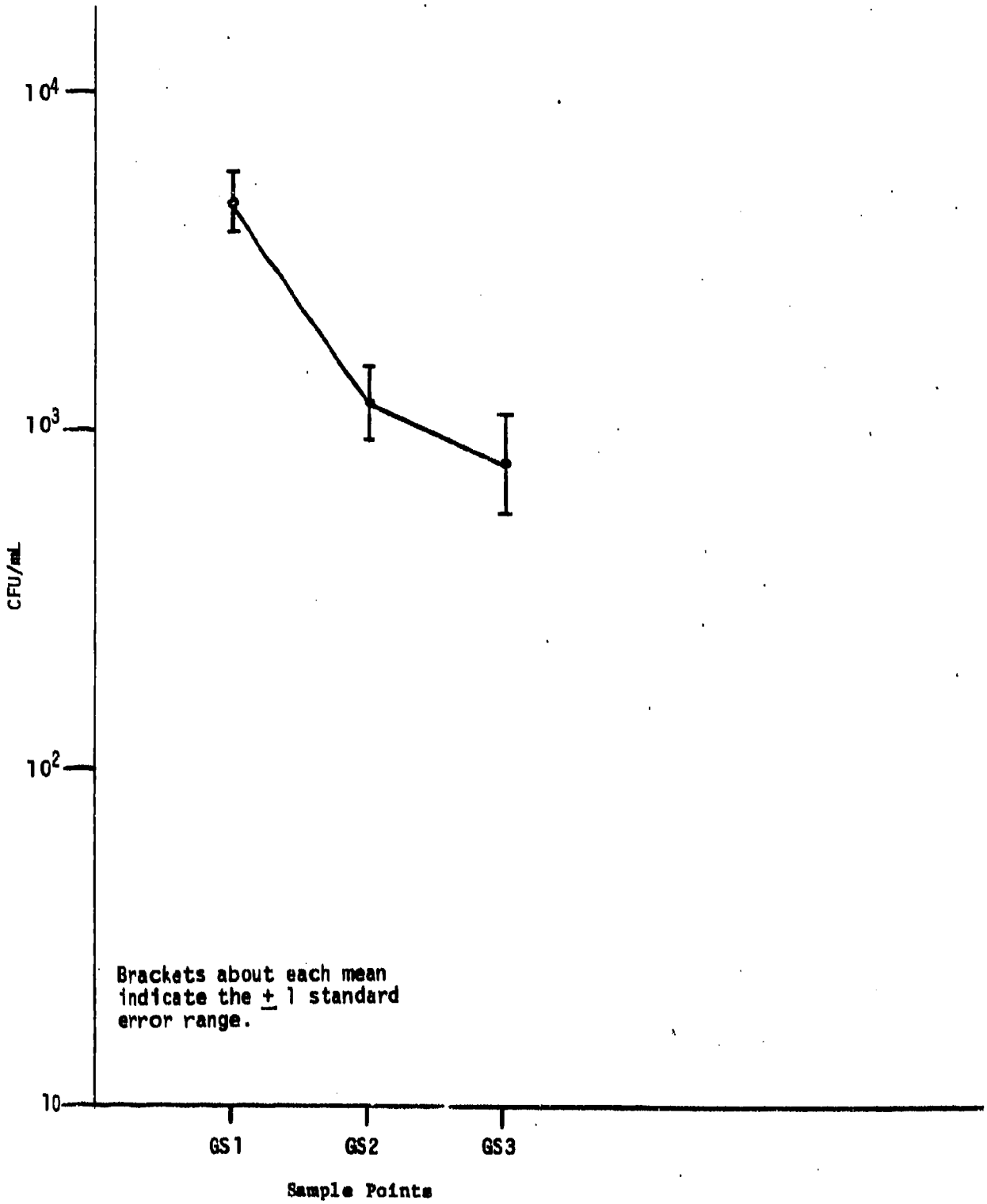


Figure 12. Summary graph: Mean total count reduction (flow rates pooled) through prefiltration system.

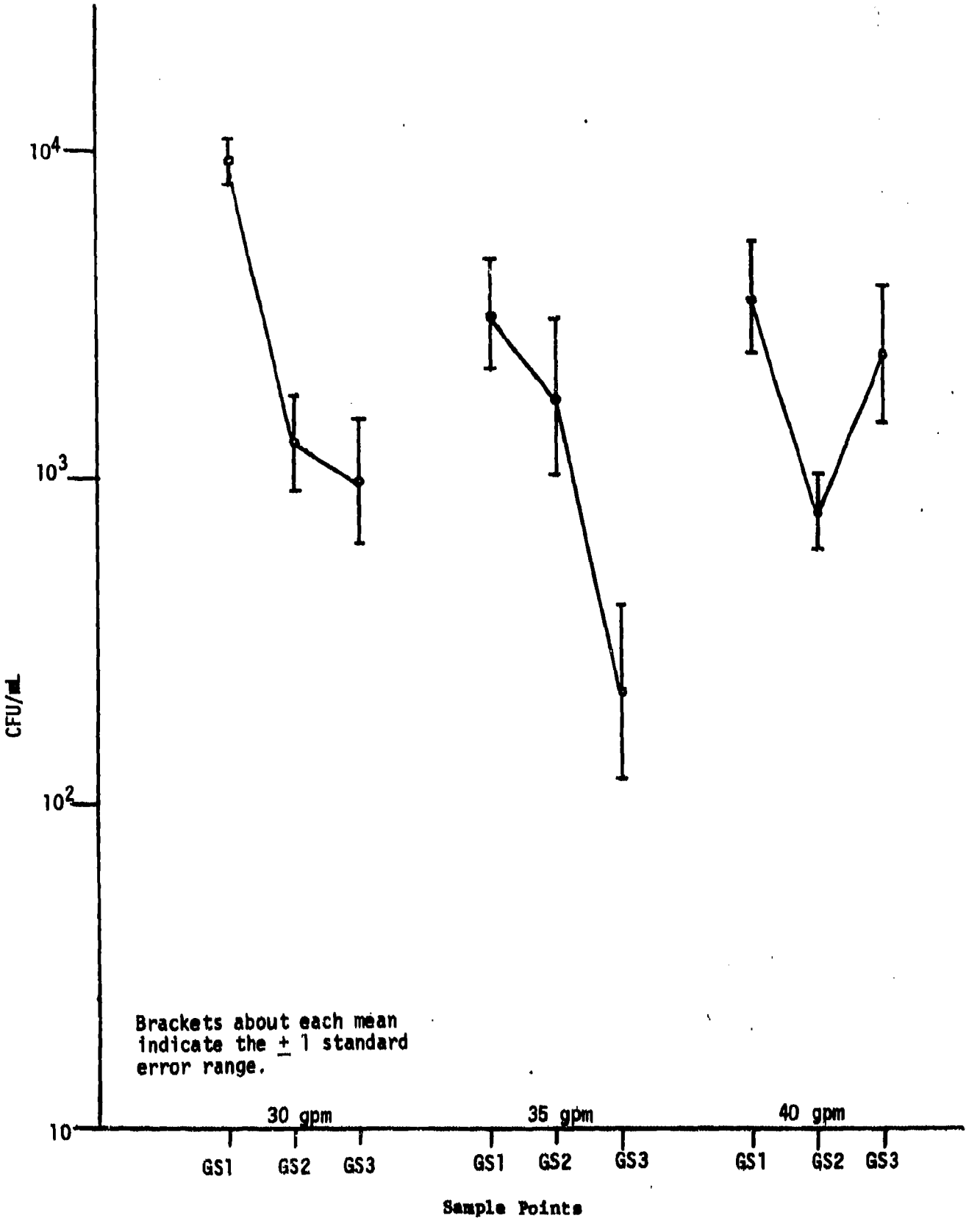


Figure 13. Mean total count reduction through prefiltration system by flow rate.

The removal performances of the MMF and CF at 35 gpm tended to compensate each other, so that across system performance, as indicated by percent removals, was somewhat better than the other flow rates. Two sets of assay data from 18 September heavily contributed to this outcome.* Examination of contemporary assays of BG, EC, and total enterics fails to show a similar trend from GS1 to GS3, which suggests the bacteria involved were background nonenterics.**

The review of data pertaining to poor CF removal performance at 40 gpm disclosed four noteworthy sequences; these are in Table 9. The sub-groups background nonenterics and non-EC enterics are roughly estimated to show which, if any, sub-group is being excessively passed. The results would indicate that the passage occurs for all groups. The breakthrough effect discussed with BG and EC thus appears to occur with these undesignated bacteria.

The total enterics ANOVA lacks the first block of data, (see Appendix C-3); hence, the impact of the 23 Sep set of data is missing. The graphical representation of the log-mean sample means are in Figure 14 (summary) and Figure 15 (flow-partitioned). The data for these graphs are in Appendix C-6.

Operational Implications

This section addresses the hypothetical configuration of the ROWPU in the production of freshwater, but with bypass of the reverse osmosis unit. This study addressed one portion of this configuration, the performance of the MMF and CF for mechanical removal of microorganisms. The study was performed during a low-river flow period, which is typical of a dry late-summer, early-fall time frame. The Monocacy River was more polluted in terms of enteric bacteria than would be considered desirable. For example, the raw water criterion for fecal coliform is a log-mean of 20 CFU/mL;[12] the *E. coli* assays at GS1 were always above that level. However, the water was not so polluted as to cause nearby conventional water treatment plants on the river to resort to unusual procedures to remove microorganisms.

* These sets are, with 35 gpm mean assays (back-transformed from log-means in Table C-5):

Time	GS1	GS2	GS3
t = 3	9,880	34,320	<100
t = 6	18,500	55,700	4,680
35 gpm mean	3,160	1,750	220

** By substracting contemporary BG and total enterics CFU/mL from the total count assay, a crude estimate of such bacteria is formed. A crude estimate of non-EC enteric bacteria can be derived by subtracting the EC CFU/mL from the total enteric assay.

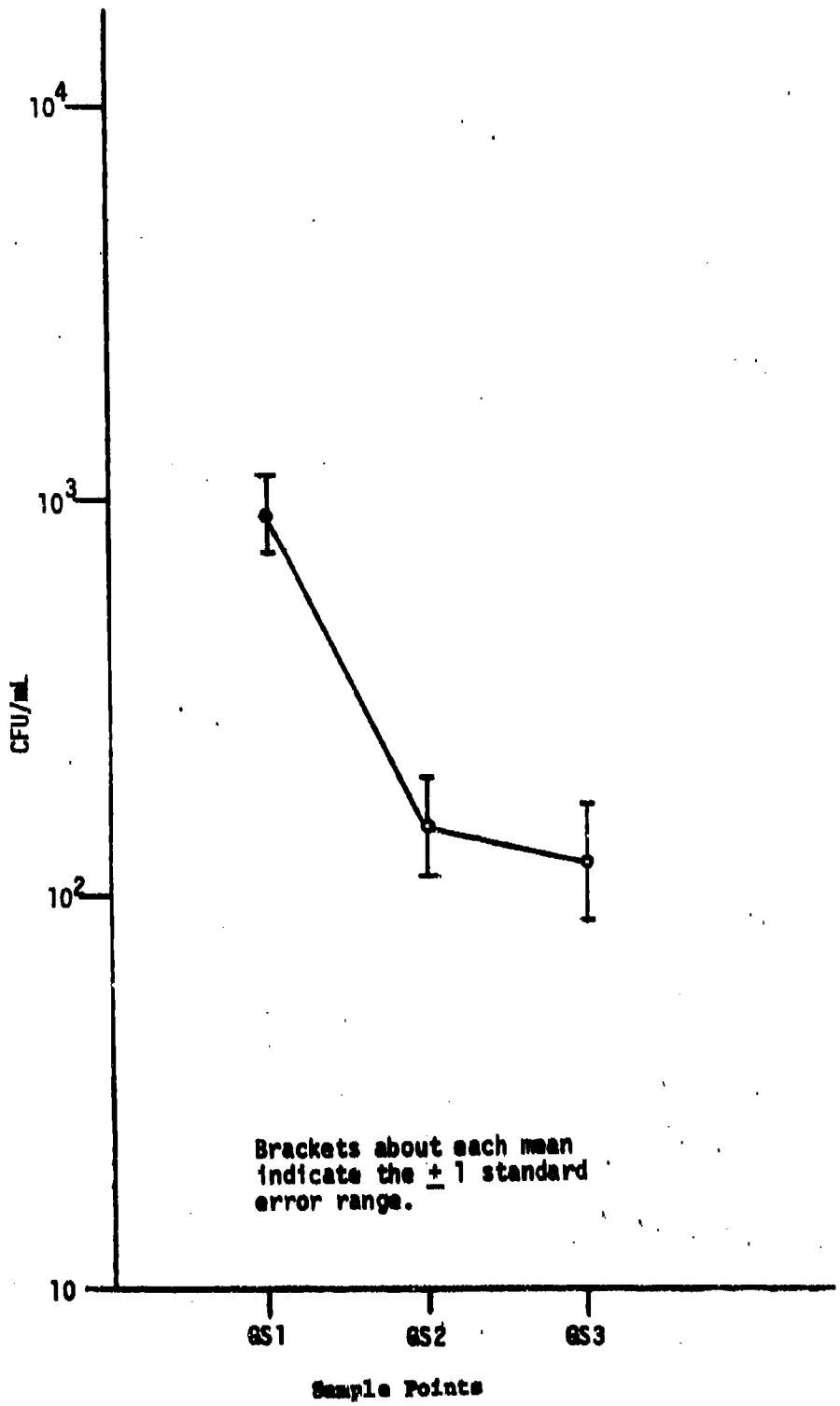


Figure 14. Summary graph: Mean total enteric reduction (flow rates pooled) through prefiltration system.

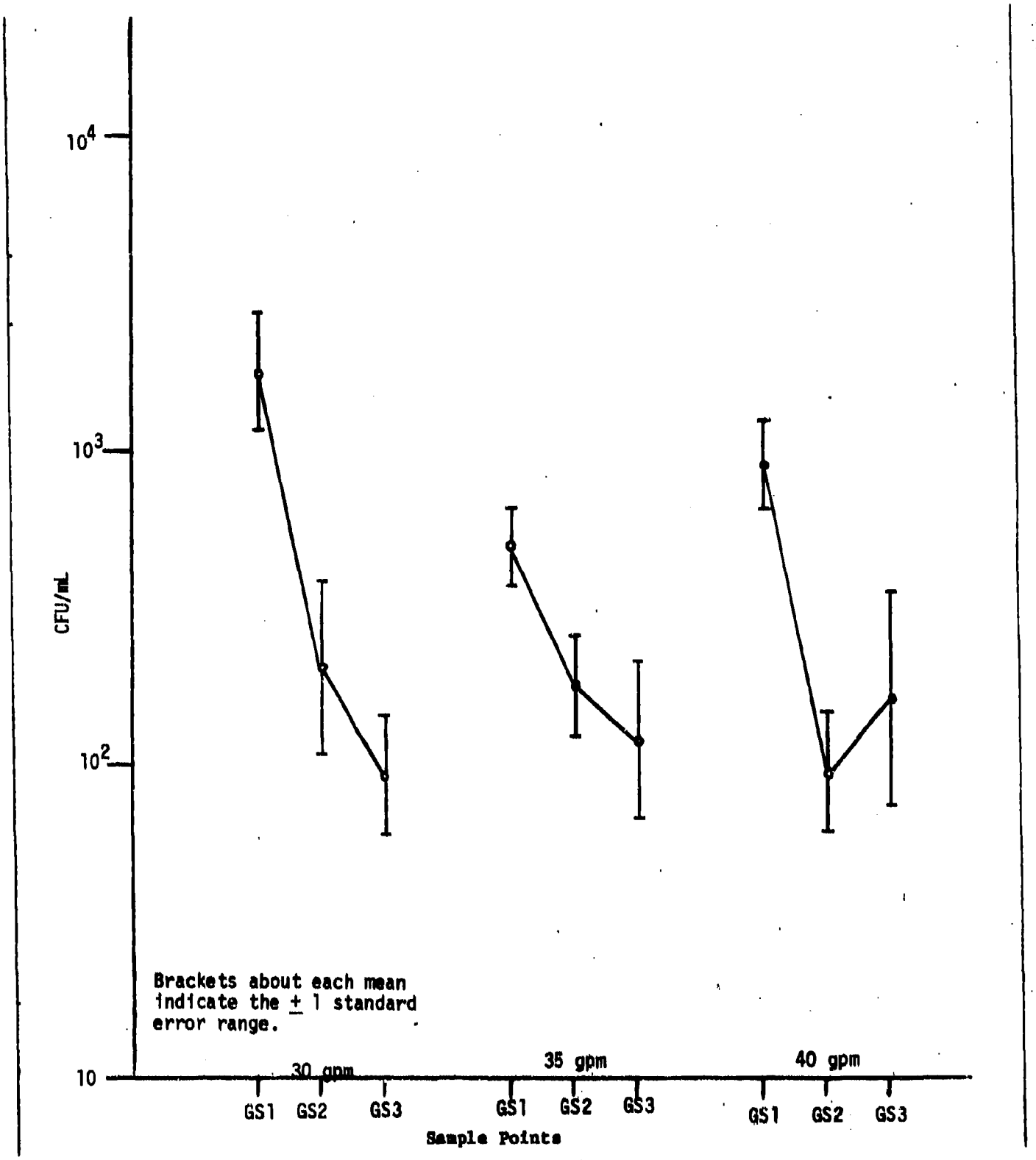


Figure 15. Mean total enterics reduction through prefiltration system by flow rate.

TABLE 9. TOTAL COUNT AND RELATED GROUPS INFLUENCING POOR CARTRIDGE FILTER REMOVAL AT 40 gpm

Time	Group	Sample Point GS1	GS2	GS3
			CFU/mL	
23 Sep, t = 0	Total count	24,290	2,230	15,400
	Background nonenterics	9,800	800	6,100
	Total enterics	11,190	1,220	8,520
	Non-EC enterics	3,900	1,100	7,300
23 Sep, t = 3	Total count	13,140	7,020	68,600
	Background nonenterics	[a]		
	Total enterics	8,160	1,110	76,390
	Non-EC enterics	7,800	1,100	76,000
23 Sep, t = 6	Total count	6,340	789	17,470
	Background nonenterics	3,500	600	4,700
	Total enterics	1,410	145	12,090
	Non-EC enterics	1,100	100	12,090
23 Oct, t = 0	Total count	1,078	848	8,646
	Background nonenterics	Nil[a]	600	1,200
	Total enterics	364	143	7,386
	Non-EC enterics	200	100	7,300[b]

a. Not determined, subtracted CFU/mL exceed total count.
b. EC data missing, GS3 assay assumed that of GS2.

The prefiltration system removal of specific organisms approached 99 percent for BG and was lower for PV, EC, and the bacterial groups. This probably reflects the physical configuration of the organisms in river water. BG, as mentioned previously, is associated with dry medium, and the physical removal depends upon the size characteristics of the medium rather than of the spores. The other bacteria and PV are probably either unattached or associated with colloid-sized particles of smaller size than BG medium. In terms of conventional water treatment, EC and the indigenous bacterial groups reflect real-world conditions. In terms of a biological warfare environment, the BG results would be a valid indicator of the removal of a dry-form organism from a water source.

The prefiltration system, by virtue of mechanical removal, would probably not be adequate in producing a product water of potable quality. Thus, disinfection would be the major line of defense against pathogens in processed water. This could pose problems in terms of the current chemical pump unit. With normal ROWPU operations, it is designed to dispense chlorine (from calcium hypochlorite) to a 10 gpm flow stream. At starting conditions suggested in the Technical Manual,[6] the dispensed chlorine corresponds to an

initial free available chlorine concentration of 8.4 mg/L. The water so treated has been processed through both filters and the reverse osmosis unit, and much of the material that exerts a chlorine demand has been removed. In this study, for example, the prefilters alone reduced demand from a mean of 1.42 mg/L to 0.89 mg/L.

In the hypothetical system, 30 gpm of water are being processed, three times that of the normal ROWPU product. The water may have considerably more chlorine demand than in the ROWPU product. The disinfectant would have to meet this demand over and above that needed for microorganism treatment.

For biological warfare environments, BG is considered a simulant for disinfection requirements. In bench-scale tests performed to establish the experimental operation procedures, an initial free chlorine level of 20 mg/L was needed to reduce BG content about 3.2 logs in 30 minutes.* While this result is not directly translatable to the hypothetical system (not enough was known about mixing regimes), it could point to a 250 percent higher chlorine level in a biological-warfare environment than projected for conventional operations, without taking into account chlorine demand.

The results of this study cannot be used to predict the system's removal of amoebic cysts. One would suspect that the cysts (about 15 micron size) are better retained than the bacteria used (about 3 micron size), but how much better cannot be determined. Further study is needed. However, if one is willing to disinfect with this prefiltration system to handle a biological warfare situation, the cysts would not be a problem (compare 20 mg/L at pH 7.4 and 22°C to a 3 mg/L residual for cyst control at pH 7.4 and 22°C).[11]

In short, the prefiltration system should not be considered a prime remover of microorganisms. Disinfection will have to do this task. If the logistical and operational considerations indicate that the disinfectant requirements are unacceptable with the present system, a change in prefiltration components may have to be considered.

The study's objective included operations at flow rates above 30 gpm, since there was interest in using larger-sized developmental units at a higher loading rate than in this ROWPU. There is somewhat poorer removal at 40 gpm than at the other flow rates. The statistical evidence for this is weak, but based on single observations, the evidence is quite striking. In terms of usual operations, where the reverse osmosis unit is to be protected against miroorganisms, avoidance of 40 gpm flow operations is suggested. Operations at 35 gpm may be acceptable; this is at best tentative, based on the restricted raw water turbidity conditions encountered.

* Distilled water at pH 7.4 and 22°C was used. At 120 minutes, the cessation of testing, a 14.6 mg/L free available chlorine level remained.

CONCLUSIONS AND RECOMMENDATIONS

The prefiltration system of the ROWPU was used to process Monocacy River water for nine separate test sessions, each lasting 9 hours. The operating conditions were designed to conform with those expected in field operations. The system was not stressed with a high turbidity water during this series of tests.

The prefiltration system mechanically removed (on a mean basis) 98.3 percent of an influent BG challenge, 93.5 percent of EC, 80.3 percent of PV, 83.1 percent of total aerobic bacteria and 86.8 percent of total enteric bacteria. This system, under the test conditions, was not a meaningful barrier to these microorganisms. These observations and their statistical significance, in terms of proposed freshwater ROWPU use, indicated that a spring 1981 test session was not necessary.

Considerable variability of removal was observed throughout the test period for the system and its component filters. The major unit process for removal was the multimedia filter. The cartridge filter exhibited unexpectedly poor removal performance at 40 gpm, possibly due to microorganism breakthrough.

Further studies should be directed to full process evaluation; that is, mechanical removal plus disinfection. Influent water quality should be adjusted, particularly for turbidity, so that the filters are hydraulically stressed. A simulant microorganism or inert material with size characteristics comparable to undesirable amoebic cysts should be used as a challenge water constituent.

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APPENDIX A

SPECIFIC EQUIPMENT USED IN ROWPU PREFILTRATION SYSTEM

Identification	Nomenclature and Manufacturer	Remarks
RW Pump #1	Burks 320WA6, 3 hp Decatur Pump Company, Decatur, IL	Standard 600 gph ROWPU pump
RW Pump #2	Barnes US4CCE, 2 hp Peabody Barnes, Inc., Mansfield, OH	Standard 420 gph "Erdulator" pump
Seed Pump	FMI Model RP-D, Serial No. B12200 Fluid Metering, Inc., Oyster Bay, NY	
In Line Mixer	Model X015-080-PVC-004-22 Komax Systems, Inc., Long Beach, CA	
Chemical Feed Pump	American Lewa FL3 American Lewa, Inc., Farmingham, MA	3-Port positive displacement pump
Multimedia Filter	Culligan MD-30 Culligan International Company, Chicago, IL	
Cartridge Filter Body	Filterite 18 FM 03A-2 Filterite Corporation, Timonium, MD	
Cartridge Filter	Filterite U005AW30A-EC1A Filterite Corporation, Timonium, MD	Six required per cartridge body
Flow Meter	Fischer-Porter 10A1755398P Fischer & Porter, Warminster, PA	0-60 gpm scale
Distribution Pump	Rex Chain Belt XP188, 2 hp REX Chain Belt Company, Milwaukee, WI	
Backwash Pump	Ampco 2 1/2 x 2ZC2, 5 hp Ampco Corporation, Milwaukee, WI	
Pressure Gages	Ashcroft S-100, 1/4-inch MPT Sockets	
Polymer	Catfloc-T, Calgon Corp. Pittsburgh, PA	
Zeta Plus Cuno Filter	Model 45144-01-1 MDS, AMF/Cuno, Maridian, CT.	

APPENDIX B

OPERATOR'S INSTRUCTIONS FOR ROWPU PREFILTRATION SYSTEM TESTS

PREFACE

These instructions will guide the test equipment operator during tests at Fort Detrick. To assure a properly conducted field test, operators must be thoroughly drilled in following them. These instructions will refer to the ROWPU Operator's and Organizational Support Maintenance Manual (Technical Manual).*

CAUTION: Operators working in the vicinity of raw water pump #1 should wear safety shoes with rippled soles for traction. Hip boots may be needed for moving intake lines.

CAUTION: Goggles and gloves should be worn when dispensing bleach; 5 percent bleach solution can irritate exposed skin and damage eyes. When using calcium hypochlorite powder, follow instructions on container. A supply of water should be available to flush any skin or eye areas exposed to this solution. Similar care should be taken with the sulfuric acid solution used for TOC sample preparation. Foul weather gear should be provided; operators may be required to work in wet weather.

I. EXPERIMENTAL SET-UP

Figure 2 of the main report is a sketch of the test equipment layout for the Fort Detrick Sewage Treatment Plant (FDSTP).

Water is drawn from the Monocacy River into the intake strainer, which is held in place by two guy lines. The guy lines are secured to trees to keep the strainer in place and to allow it to be maneuvered from above. The strainer is attached to 75 ft of 2.0-inch ID plastic hose, which is attached to raw water pump #1.

The source of water was selected to provide a reasonably accessible position along the bluff on which the FDSTP is located and to still meet the head/suction limitations of the pumps. Consideration was given to the possibility of floods. Raw water (RW) pump #1 is about 5 ft above the mean flow water level. Heavy rains may raise water levels sufficiently to submerge this pump. To prevent water damage to this pump, a hoist is provided to raise the pump up the hill.

Water pumped from raw water pump #1 flows through a 110-ft section of 1.5-inch polyethylene tube or pipe to raw water pump #2. Water pumped from raw water pump #2 flows through a 45-ft section of 2.0-inch polyethylene tube to the test equipment (starting at the RW tap).

* U.S. Army Mobility Equipment Research and Development Command, Fort Belvoir, VA. 1979. Operators and organization support maintenance manual for 600 gph reverse osmosis water purification unit (DRAFT).

The flow exiting the test equipment is chlorinated. Then a 15-ft section of hose is used to route product water to the top of a 1,500-gal holding tank (HT). Water exits this tank through the HT valve. During water purification, flow is directed to the FDSTP chlorination tank for disposal. During backwash, flow is directed to the backwash pump and through the backwash gate valve. The multimedia filter has a timer/valve control which performs the backwash sequence. Spent water flows out of the backwash drain valve and to a FDSTP sand bed for disposal.

II. WEEKLY START-UP PROCEDURES

1. Prior to the start of a 3-day test block, sufficient supplies will be delivered to the test site from USAMBRDL. Required items are listed in Section IV, Table 1. This list should be used as an inventory to ensure that no items are missing.

2. If necessary, lower raw water pump #1 to its base with the hoist. Connect the water intake line to the suction-side union of the pump. Connect the line to raw water pump #2 to the discharge-side union of the pump. With guy lines, maneuver the strainer intake to a good position in the river.

3. If system is in "weekly shutdown condition," place all valves in "prior to start" position (see Section [illegible] Table 2).

4. Place all valves in "prior to start" position (see Table 2).

III. DAILY OPERATIONS

Samples will be labeled using the format (Julian date)-(Tap)-(Time)-(Analysis). A sample assay request sheet is needed for chemical assay samples.

1. Prior to each day's test, expected river level behavior will be estimated. Reposition strainer intake with guy lines for the planned daily intake point.

2. Verify that equipment is ready for testing:

   A. All water lines are connected as indicated in flow diagram (Figure 2, main text).

   B. The power to all pumps is off and all pumps are properly plugged into power supply.

   C. Valves settings are in the "prior to start" position (see Table 2).

3. Seed suspensions will be picked up at USAMBRDL prior to each day's test. Keep cool until added to Bio Seed Tank. Fill this tank to 10-gal level with tap water. Stir for 5 min and then determine chlorine residual level (see Step 23C). If residual exists, add 1 mL of 10 percent sodium thiosulfate solution per 1 ppm of residual. Stir again for 5 min and measure residual again. If residual still exists, add 0.2 mL of sodium

thiosulfate solution. Repeat stirring, residual determination, and thiosulfate addition until no residual is noted.

4. Prepare chemical solutions as follows:

   A. Polymer: Add 110 mL polymer to 3 gal water for 30 gpm; 125 mL polymer for 35 gpm; and 140 mL polymer for 40 gpm.

   B. Bleach: For 30 gpm flow, dilute 7 qt bleach to 3 gal with water; for 35 gpm flow, dilute 8 qt bleach to 3 gal with water; for 40 gpm flow, dilute 9 qt bleach to 3 gal with water.

   CAUTION: Do not cross-contaminate chemicals by splashing.

5. Place polymer pail under port 3 of chemical pump and bleach pail under port 2 of chemical pump. Inlet lines must be primed; use tap water. Turn on pump and set port flows at 3.0. Check flow rate from return lines by measuring flow collected in 1 min, once for each port. Acceptable flows are 57 to 63 mL/min. Enter calibration data on daily log sheet. Have return lines drain to pails when complete. Adjust port flow knob 0.1 units for each 2-mL change in flow desired.

6. Prime raw water pump #1 with water from the prime tank. Make sure pump is filled to brim. Do this immediately before Step 7. Note: Flow of water from raw water pump #1 will prime raw water pump #2.

7. Turn on raw water pump #1. Wait 15 seconds and open line bleed valve. When air is vented from line bleed valve and water gushes out, close it. This pump should suffice to carry water to process. When steady stream of water passes through RW and GS #1 valves, close them. Open (FLOW REG) valve. When steady stream of water passes through multimedia filter (DF) vent, close it. Open cartridge filter (CF) valve. When steady stream of water passes through CF vent valve, close this valve.

8. Turn the polymer return valve off and the feed valve on. Note: Always close return valve fully before opening feed valve.

9. Adjust water flow to desired setting. Record time of this event on log sheet. RW tap may be kept open to relieve pressure on pumps and augment flow control.

10. Add seed suspensions to Bio Seed Tank. Record time of this event on log sheet.

11. Perform clarity check and adjust polymer flow, if necessary, according to Technical Manual, paragraph 5.1.B.(7). Record flow setting for clear water on log sheet. Allow at least 10 min between Step 9 and Step 11.

12. Turn bleach return valve off and feed 1 valve on.

13. After seed check valve is holding water, connect seed suspension line to swaglock fitting. After at least 20 min has elapsed from Step 10, turn on seed pump. Note time of this event on log sheet.

14. Open holding tank valve and turn on distribution pump. Notify FDSTP personnel that discharge to their system has begun. Check tank level. Proper behavior is constant or slowly rising level.

Throughout test, the holding tank valve may have to be adjusted to prevent overflow or the water level dropping too low. Ideally, the level of water should be in the 1,000 to 1,400-gal range. Distribution pump will handle 45 gpm.

While the following steps are sequential in narrative, the operator will have to devise a routine to do them in a minimal amount of time. Clean-up and disposal instructions are given in Step 22.

15. Starting with the RW tap, follow these steps:

    A. Collect 2 liters of water in a 2-liter beaker.

    B. Insert thermometer in beaker and record temperature.

    C. Read multimedia filter inlet and outlet pressures.

    D. Read cartridge filter outlet gage.

    E. Read flow rate and adjust if necessary.

    F. Record time and data from Steps B, C, D, and E on log sheet.

16. RW sample: Fill a 1-liter polypropylene bottle with sample from RW tap. Label bottle: (Julian day)-RW-start-EC/BG.

17. Seed tank sampling procedure:

    Pipette 9 mL of seed suspension into a test tube which has 1-mL of Hank's balanced salt solution. Label tube: (Julian day)-seed-start-PV. Pipette 10 mL of seed suspension into a second test tube. Label tube: (Julian day)-seed-start-EC/BG.

18. Place sample collected from Steps 16 and 17 under refrigeration in the Bio chest.

19. GS1 tap sample procedure:

    A. Collect 2 liters of sample in a 2-liter beaker.

    B. Fill a 1-liter polypropylene bottle with sample from the GS1 tap. Label bottle: (Julian day)-GS1-start-EC/BG.

    C1. Direct Virus Sample. Pipette 9 mL of sample into a screw-cap test tube which has 1 mL of Hank's Balanced Salt Solution. Label tube: (Julian day)-GS1-start-PV.

C2. Concentrated Virus Sample (6 hr sample only). Process water from GS1 tap through virus concentration apparatus (see Fig. 4, main text). Instructions are at step 24.

D. Transfer 250 mL of sample to a 250-mL polyethylene bottle. Label bottle: (Julian day)-GS1-start-T/A.

E. Transfer 250 mL of sample to a 250-mL polyethylene bottle. Pipette in 2 mL of sulfuric acid solution. Screw on cap and shake. Label bottle: (Julian day)-GS1-start-TOC.

F. Place samples from Steps B-E in chemical or bio ice chests as appropriate.

G. Retain remainder of sample for on-site analysis (Step 23).

20. GS2 tap sample procedure:

A. Wait at least 10 minutes from step 19A.

B. Collect 2 liters of sample from tap in a 2-liter beaker.

C. Fill a 1-liter polypropylene bottle with sample from the GS2 tap. Label bottle: (Julian day)-GS2-start-EC/BG. Place in bio ice chest.

D. Transfer 250 mL of sample to a 250-mL polyethylene bottle. Label bottle: (Julian day)-GS2-start-Turb. Place in chemical ice chest.

E. Process water from GS2 tap through virus concentration apparatus.*

21. GS3 tap sample procedure:

A. Collect 2 liters of sample from tap in beaker.

B. Follow Step 19B, except that GS3 rather than GS1 designation is on label.

C. Follow Steps 19D and 19E, except that GS3 designation is on label.

D. Keep remainder of sample in 2-liter beaker for on-site analysis (Step 23).

E. Process water from GS3 tap through virus concentration apparatus.*

22. Clean-up procedures and good lab practices during run:

A. Label all beakers and pipettes to avoid mix-up.

* Instructions were written before decision was made to direct-sample these taps. For such samples, Step 19C1 applies with proper sample point labelling.

B. Rinse out sample beakers with new sample prior to collecting sample for record.

C. Dump unused samples and rinse waters into 3-gal wastewater storage buckets (see Step 31C).

D. Pipettes, test tubes, and bottles used in chlorine demand analyses should be used only once. Pipettes should be discarded and test tubes and bottles should be placed in a 3-gal bucket with chlorinated product water after use.

23. On-site analysis:

A. pH: Expose air hole, then insert pH probe in sample. Swirl probe and turn dial to pH setting. Swirl for at least 30 sec. Read valve off chart. At the completion of each measurement, rinse off electrode with distilled water and shake dry. Record measurement on daily log sheet.

B. Total dissolved solids: Rinse sample cup with water to be tested. Discard. Pour in sample of water and read meter. Discard this sample. Add second sample and read meter. Discard sample. Record both readings on daily log sheet. Record reading for 200 mg/L salt solution. Rinse cup with distilled water.

C. Chlorine demand test: Add 100 mL of sample to each of three 125-mL polyethylene bottles. To one bottle, add 0.2 mL of test bleach solution. To the second bottle, add 0.4 mL of test bleach solution. Add nothing to the third bottle. Stopper and shake each bottle for 10 sec. After 20 min, perform FACTS test on each bottle as follows:

(1) Add 5 mL of sample to test tube.

(2) Add 0.2 mL FACTS buffer solution.

(3) Add 2 mL FACTS reagent.

(4) Stopper test tube and invert twice.

(5) Compare sample color to standards. Read estimated concentration within 30 sec. Estimate either to the closest concentration to the color or the bracket in which the color falls. Record concentration in appropriate section on daily log sheet.

River conditions may influence chlorine demand. If initial bleach additions cause excessively high FAC readings, repeat test with lower test bleach concentrations. Conversely, if test bleach concentrations are not sufficient to cause a residual, raise test bleach concentrations. Tests may be reduced to two bottles (one with bleach and one without) for sample times after the "start" sample.

24. Virus concentration procedures:

A. Open filter element plastic bag or paper wrapper carefully.

B. Insert filter element on filter rod and screw tight.

C. Place filter element and rod in housing. Insert rod with filter and screw on housing.

D. Raise drain line to level above top of spent sample collection tank (provided with concentration apparatus). Add 25 mL of bleach solution from chemical feed bucket to tank.

E. Open virus sample line on tap. Simultaneously open regulation valve on filter assembly and press red vent button. Adjust flow to 1-2 gpm.

F. Allow filter to process 200 liters of water as measured by tank level. Close regulation valve and line sample tap.

G. Remove filter element and shake off excess water. Add 600 mL of nutrient solution to container provided and place filter element in container. Cover container and refrigerate.

H. Lower drain line and allow tank to drain. Replace filter housing.

25. Hourly readings:

A. Repeat procedure of Step 15.

B. After use, discard raw water sample collected.

C. If hourly readings conflict with Steps 16-21, take readings immediately prior to these steps.

26. Samples collected at 3 and 6 hours after startup:

A. Repeat Steps 17-21.

B. Label samples as 3 hour or 6 hour in appropriate time slot.

27. Sample collected at shutdown (start +9 hours):

A. Repeat Steps 16-21.

B. Label sample as 9 hour in appropriate time slot.

28. Every 3 hr, it will be necessary to prepare polymer and bleach solutions and add to pails. Don't let hoses run dry.

29. Preparation for backwash: After samples have been collected:

A. Shut off distribution pump (if used) and close holding tank and back-up drain valves.

B. Shut off seed pump C. As holding tank reaches top, shut off raw water pump #2 and raw water pump #1, in that order.

C. Close CF valve and regulation valve.

D. Turn polymer feed/return valves to off/on positions. Turn polymer pump setting to 0.0.

E. Turn bleach feed 1/feed 2/return valves to off/off/on position. Set port 2 flow setting to 10. Make sure there are at least 2 gal of bleach chemical at this time; otherwise, prepare sufficient chemical for this volume.

F. Add 2.5 liters of 10 percent sodium thiosulfate solution to holding tank.

30. Backwash instruction:

A. Open backwash drain valve.

B. Turn on backwash timer. If necessary, turn cams until black cam (not the bottom cam on the four-cam train) is almost engaged.

C. Turn bleach valves feed 1/feed 2/return to off/on/off position.

D. Turn on backwash pump.

E. When black cam engages, open backwash gate valve.

F. About 4 min should pass before backwash starts. If cams do not appear to move, push "extra recharge" lever.

G. Backwash is completed when the fourth cam on the train disengages. Shut off timer, close backwash (BW) gate valve, and shut off pump. Close backwash drain valve.

H. Turn bleach valves feed 1/feed 2/return to off/off/on.

31. Daily shutdown (assume a run the next day):

A. Instruction will be given on position of strainer inlet line until next test.

B. Seed tank and pump. Remove seed intake line from tank. Add 100 mL of bleach solution from chemical feed bucket to seed tank. Continue stirring for 10 min, then dump tank contents on sand bed. Flush out seed tank with tap water. Meanwhile, add 2 liters tap water and 5 mL bleach solution from chemical feed bucket to a 2-liter beaker. Remove seed line from swaglock fitting. Insert inlet of seed line into beaker and allow pump to run 10 min, draining into a wastewater bucket. Discard residual water in beaker. Add 2 liters tap water and 2 mL sodium thiosulfate solution to beaker. Repeat pumpout for 10 min. Discard residual water in beaker.

C. Add 100 mL of bleach solution from chemical feed bucket into any wastewater buckets. Stir, wait 30 min, and dump waste buckets in sand bed.

D. Wash out polymer bucket with tap or collected production water. Dump on sand bed.

E. Return all valves to "prior to start" position.

F. Drain test tubes and 125 mL bottles from FACTS tets. Place in plastic bags. Dump water in pail on sand bed.

G. Discard any bleach from chemical feed bucket in sand bed. Flush out bucket with tap water.

H. Return all samples to lab.

32. Special shutdown procedures:

A. Plugged multimedia filter: A multimedia filter is considered plugged if the pressure drop at any time in the test exceeds the start-up pressure drop by 5 psi. If this happens, note time on log sheet and immediately (as soon as it is practical) collect samples according to Step 27. Then proceed for backwash and shutdown.

B. Plugged cartridge filter: Filter is considered plugged if pressure drop reaches 25 psi or flow through system cannot be maintained. If either of these happens, note time on log sheet. Proceed for backwash and shutdown. Cartridges will have to be replaced before next test.

C. Emergency move for raw water pump #1:

(1) Open prime bleed. Allow water to drain.

(2) Break unions to inlet line and line to raw water pump #2.

(3) If conditions allow, move strainer line to higher ground and secure it to a tree with rope.

(4) Move pump partway up hill with winch attached to tree. One person will have to guide skid and the other person will pull. Watch for snagging ropes on power line.

(5) Tighten rope from tree to pump and release winch jaws. Reconnect jaws to U-bolt on rope at top of hill (this rope is connected to fence post).

(6) Place block between lower end of skid and rung on ramp. Loosen rope from tree. While one person holds rope in light tension, others transfer skid from ramp that goes partway up hill to ramp which goes to hill crest.

(7) Repeat Step (4) with ramp to hill crest.

(8) Secure at hill crest or move pump to higher ground manually or with a pushcart.

D. Freezing weather: If freezing temperatures are expected, follow these steps:

(1) Drain prime case on raw water pump #1.

(2) Open line bleed valve.

(3) Drain prime case on raw water pump #2.

(4) Break union between RW tap and inlet test section.

(5) Place system in "weekly shutdown" mode.

(6) Remove outlet hoses from chemical feed pump and allow pumps to run 5 min at a 10.0 setting to displace water.

(7) Break union between holding tank and backwash pump.

(8) Remove hose from backwash pump to multimedia filter inlet tee.

(9) Drain prime case on distribution pump.

(10) Allow RW #1, RW #2, and distribution pumps to run for about 2 min each to fling off water.

33. Special procedures:

Testing may be done in rainy weather. Plan to take samples inside at chlorine building for analysis preparation and packaging. Provision will have to be made to batten down items during windy weather or thunderstorms. Generally, runs will not be done during periods of rapidly rising water, as riverborne debris may wash away lines. Moreover, runs may be postponed in periods of rapid river stage changes, such as immediately after heavy rains.

34. Weekly shutdown and maintenance:

A. Perform daily shutdown procedures of Step 31 if not completed.

B. Place valves in "weekly shutdown" position. This action will drain the multimedia filter via the auxiliary drain valve and the cartridge filter via the floor plug.

C. Perform chemical-pump cleanout as described in Technical Manual paragraph 5.8.B.(4C-I).

D. Perform periodic maintenance of equipment as described in the following sections of the Technical Manual: Table 8-1, items 1, 3, 5, 7, 8, 9, 11, 13, 16, 17, 19, 20, 21, 24, and 25.

E. Store all buckets, empty tanks, and equipment in storage area.

F. Transport all plastic bags with used chemical bottles, biological flasks, and used filter to laboratory for disposal or cleanup. Also transport back any undelivered samples.

G. Bleach preparation: Add 300 mL of calcium hypochlorite pellets to an empty 1-gal bleach bottle. Fill jug with 1 gal tap water or product water, close lid, and shake for 30 sec.

H. Glassware cleanup:

(1) One-liter polypropylene bottles should be autoclaved by microbiological personnel and returned for reuse.

(2) Screw-cap test tubes should be returned to microbiological personnel for autoclave or disposal.

(3) Two hundred fifty-milliliter polyethylene bottles for chemical samples should be washed with 15 ppm bleach, rinsed, washed with distilled water, and air dried.

(4) Test tubes and bottles for FACTS tests should be returned to Building 459 at end of test week. They should be rinsed in distilled water, shaken dry, rinsed in demand-free water, and air dried.

I. During down time, clean bottom of holding tank with squeegee or wet vacuum.

J. Test bleach solution should be prepared prior to the start of each 3-day block. Test solution is 4 mL of Clorox brand bleach to 196 mL of distilled water.

E. Store all buckets, empty tanks, and equipment in storage area.

F. Transport all plastic bags with used chemical bottles, biological flasks, and used filter to laboratory for disposal or cleanup. Also transport back any undelivered samples.

G. Bleach preparation: Add 300 mL of calcium hypochlorite pellets to an empty 1-gal bleach bottle. Fill jug with 1 gal tap water or product water, close lid, and shake for 30 sec.

H. Glassware cleanup:

(1) One-liter polypropylene bottles should be autoclaved by microbiological personnel and returned for reuse.

(2) Screw-cap test tubes should be returned to microbiological personnel for autoclave or disposal.

(3) Two hundred fifty-milliliter polyethylene bottles for chemical samples should be washed with 15 ppm bleach, rinsed, washed with distilled water, and air dried.

(4) Test tubes and bottles for FACTS tests should be returned to Building 459 at end of test week. They should be rinsed in distilled water, shaken dry, rinsed in demand-free water, and air dried.

I. During down time, clean bottom of holding tank with squeegee or wet vacuum.

J. Test bleach solution should be prepared prior to the start of each 3-day block. Test solution is 4 mL of Clorox brand bleach to 196 mL of distilled water.

TABLE B-1. SUPPLIES FOR 3 DAYS OF TESTING

Chemicals

1,125 mL Catfloc-T polymer
19 gal 5% hypochlorite bleach or 12 lb technical grade calcium hypochlorite
50 mL 1 M sulfuric acid solution for TOC sample preparation
10 mL FACTS buffer solution
100 mL FACTS indicator
25 mL 1:50 Clorox/water solution for FACTS sample chlorination
8 liters 10% (W/W) sodium thiosulfate (technical grade) solution
2 gal of distilled water in carboy
600 mL of 200 mg/L sodium chloride (technical grade) calibration solution for TDS meter
7.2 liters of nutrient solution for virus concentration

Sample Containers and Incidentals

Several 3-gal pails to prime pumps, carry water, hold garbage
4 3-gal plastic pails for chemical feeds
4 2-liter glass beakers for sample collection
1 thermometer
1 2-mL automatic pipette for sulfuric acid transfer with disposable glass tubes
60 5-mL disposable pipettes for FACTS sample transfer
1 1-mL pipette for bleach solution dosing
1 nosedropper or 1 mL-pipette for FACTS buffer solution
1 nosedropper or 2-mL automatic pipette with disposable glass tubes for FACTS indicator
2 marked bottles for FACTS sample transfers
42 1-liter polypropylene bottles for Bio samples
60 250-mL polyethylene bottles for chemical samples
48 125-mL polyethylene bottles for FACTS samples
60 test tubes for FACTS analysis
36 screw-cap test tubes for seed suspension samples
1 marked bottle for polymer addition
2 ice chests, one for Bio samples and seed, one for chemical samples
1 100-mL beaker for calibration
36 10-mL disposable pipettes for seed suspension collection
1 beaker and funnel for calcium hypochlorite

TABLE B-2. VALVE SETTINGS FOR NORMAL CONDITIONS

Valve or Tap	Prior to Start	Run	Backwash	Weekly Shutdown
Line bleed	closed	closed	closed	open
Regulation	closed	adjusted	closed	open
RW	open	samples only	closed	open
GS1	open	samples only	closed	open
GS2	closed	samples only	closed	open
GS3	closed	samples only	closed	open
Polymer(feed/ return)	closed/open	open/closed	closed/open	closed/open
Bleach (feed 1/ feed 2/return	closed/closed open	open/closed closed	closed/open closed	closed/closed open
HT	closed	adjusted	closed	open
Backwash drain	closed	closed	open	open
Cartridge and multimedia filter vent	open	closed	closed	open
Backwash gate	closed	closed	open	open
Back-up drain	open	adjusted	closed	open
DF auxiliary drain	closed	closed	closed	open
CF	closed	open	closed	open
DF and CF floor drain plugs	closed	closed	closed	open, CF only

APPENDIX C

STATISTICAL APPROACH

This Appendix is subdivided into three parts. The first part discusses the specific multifactor ANOVA used in processing log (assay) data. The second part discusses, in statistical terms, the impact of curtailing operations as mentioned in the "Results and Discussion" section. The third part discusses each specific microorganism/group analysis and presents tabular information which is used in the "Microbiological Data Analysis" section.

C-1. The Multifactor Analysis of Variance.

The multifactor analysis of variance was applied to a given assay's data, which considered sample point, elapsed time, and flow rates as treatments. Treatment interactions were also investigated. The analysis is included in the Statistical Analysis System computer programs, as cited in the SAS User's Guide.* Specifically, flow rate occurs at three levels (30, 35, and 40 gpm), elapsed time at four levels (t = 0, 3, 6, and 9 hours), and sample point at three levels (GS1, GS2, GS3). The complete set of factors were replicated three times, i.e., blocks. From this design a full set of data consisted of 108 observations for each microorganism or group, 36 at each sample point, 27 at each elapsed time, and 36 at each flow rate.

The multifactor analysis model is summarized in Table C-1. The model is evaluated by testing the null hypotheses that log (assay) is not influenced by elapsed time, flow, sample point and that no interactions of these statistics exist. The ANOVA assesses the probability of whether these are valid hypotheses, using F-distribution criteria to determine probability values (p-values); see Section C-2 below. The flow and elapsed time effects and the FxT interaction are tested against their respective interactions with blocks. The sample point effect and the FxS, SxT, and FxSxT interactions are tested against the residual variance or error. This latter feature is particularly attractive since the error term is based on a large number of degrees of freedom and is free of systematic effects of block, flow, elapsed time, and these effect interactions. Thus, the error can be used for evaluation of paired sample point comparisons. The t-test for means is used, from which a p-value is determined for each pairwise comparison.

The F-tests in the ANOVA are functions of the standard errors of the means. Therefore, for the discussion presented in the main text, the standard error of the mean is presented as a measure of variability. The standard error measures the precision by which a mean is estimated from the experimental data.

* SAS Users Guide 1979 ed., SAS Institute, Inc., Raleigh, NC.

TABLE C-1. ANALYSIS OF VARIANCE MODEL

(a) For Flow and Time

Variance Component	Degrees of Freedom	Remarks
Blocks (B)	2	
Flow (F)	2	
Elapsed Time (T)	3	
FxT	6	
BxF	4	F-Test for flow
BxT	6	F-Test for time
BxFxT	12	F-Test for FxT
(b) For Sample Points(s)		
S	2	
FxS	4	
TxS	6	
FxTxS	12	
Error	48	F-Test for S, FxS, TxS, FxTxS
Total	107	

C-2. Statistical Implication of Test Schedule Curtailment

Statistical analysis can be generalized to three parameters. The first, p-value, measures the probability that the observed treatment effect is due to chance variation alone; for example, the mean log (CFU/ml) of BG change between sample points is due to chance or the imposed treatment. Statistical procedures involve selection of a second parameter, called alpha, the level of significance. The smaller alpha is, the less likely that an outcome is falsely attributed to a treatment effect when in fact (and never known to the experimenters) the outcome is due to chance. Typically, alpha values of 0.10 or 0.05 are preselected for comparison to p-values. Effects with p-values less than alpha are called "statistically significant," that is, by experimenter judgment, unlikely to be due solely to chance variation.

The last parameter is not directly addressed in a study such as this, where one seeks to determine if some effect causes a change in observation. This parameter is called beta; (1-beta) is the power of the test. The power of the test measures the probability that if a specified change did exist, the statistical analysis would indicate the change. The power of a test is important where a decision to take an alternative course of action hinges on the change being of a stated amount. The power of the test needs to be stated when commenting upon the sensitivity of a test plan.

For paired changes in S, FxS, SxT or the paired comparisons, sensitivity also involves the size of the error term degrees of freedom. The table below presents minimum differences in actual (not experimental) log means that could

have been detected for each microorganism/group at an alpha of 0.05 and (1-beta) of 0.90.

Microorganism/ Group	Experimental Error, Log Units	Detectable Change, Log Units, this Study	Detectable Change, Log Units, 8-Block Study
BG	0.4793	0.37	0.23
EC	0.5438	0.42	0.26
PV	0.3152	0.32	0.18
Total counts	0.6131	0.47	0.29
Total enterics	0.6297	0.61	0.32

For example, from Table C-3, the pooled log mean GS2 assay was 1.7118; that of GS3, 1.5835. The 0.1282 log unit difference was not, in view of experimental error, significant at alpha = 0.05. If this difference was, in fact, 0.37 log units, a 90% probability exists that this study, repeated many times, would have the outcome that the experimentally determined difference had a p-value <0.05. Had the projected 8-block test been concluded, the above statement would be valid for a 0.23 log-unit difference.

The situation with elapsed time, flow, and the FxT interaction is more complex. In this study, these effects (with the exception of the total count flow effect) were not statistically significant, so an in-depth analysis of test plan change is not done. In this case, the number of replicated blocks are involved, and the following table shows the number of blocks required to detect specific differences based on experimental error for p = 0.05, (1-beta) = 0.90.*

Experimental Error, Log Units	Replicated Blocks Required for Cited Difference 1/2 Log Unit	1 Log Unit	1.5 Log Unit
1.0	>70	22	10
0.5	22	6	4
0.33	10	5	3

In comparison to the previous table, the sensitivity change between an 3-block test and an 8-block test is much greater. For example, for a 3-block test with 0.33 log unit experimental error, 1.5 log unit differences are detected with p <0.05 90% of the time. For the 8-block test, this error corresponds to a interpolated detection limit of about 0.6 log-units. This higher loss in sensitivity was considered justifiable, in that these effects are less important to the study objectives than were sample-point associated effects.

* From Neter, J. and W. Wasserman. 1974. Applied Linear Statistical Models. Richard D. Irwin, Inc., Homewood, IL.

C-3. Specific ANOVA Details and Intermediate Results

Each microorganism or group was treated as an independent entity. The assay data (Appendix F, Tables 3-7), were transformed to log (assay), the statistic analyzed. Where qualitative data were involved, the assay result was represented as 1 CFU/mL.

The analysis for BG was on a full set of data. The p-values derived for effects are in Table 7, main text. Intermediate log-means and their standard errors are in Table C-2. The log mean sample point standard errors (pooled and flow partitioned) reflect the variance derived from the specific observations that compose each log mean. The paired comparison standard errors involve the error variance from the ANOVA, and for the full-set case, all are equal.

TABLE C-2. INTERMEDIATE ANALYSIS RESULTS CITED IN MAIN TEXT: BACILLUS GLOBIGII

(a) Log Means and Standard Errors Sample Point, Pooled and Flow-Partitioned

Sample Point	Pooled	Flow Rate, gpm		
		30	35	40
		Log Mean ± Standard Error[a]		
GS1	3.3472±0.0788	3.4195±0.1352	3.4148±0.1266	3.2072±0.1494
GS2	1.7118±0.1111	1.8381±0.1141	1.9941±0.2391	1.3032±0.1541
GS3	1.5835±0.0984	1.6684±0.1047	1.5663±0.1946	1.5158±0.2061

(b) Paired Comparisons

Paired Comparison	Log Mean ± Standard Error
GS1-GS2	1.6354±0.1130
GS1-GS3	1.7637±0.1130
GS2-GS3	0.1283±0.1130

a. Each pooled result based on 36 observations; each flow rate result on 12 observations.

The analyses for Escherichia coli had to adjust for missing data; 103 observations were available. The intermediate results are in Table C-3. Because of missing data, the paired comparison standard errors are unequal.

The sample design for PV was less thorough than for BG. GS1 was sampled at t = 6 hours, and GS2 and GS3 at t = 3 and t = 9 hours. This was due to supply limitations and the scheduling of operations during tests. In the ANOVA, the GS1 and GS3 assays transformed represent the arithmetic averaged

results at t = 3 and t = 9 hours. The ability to measure elapsed time and elapsed time-interactive effects was lost. This was considered an acceptable loss of information.

The data in Table F-5 and the intermediate mean results of Table C-4 are reported in terms of unconcentrated water samples, without adjustment for concentration efficiency. This is for consistency with other microorganism assays. The assay of record is in terms of PFU/mL of concentrate, and Figures 10 and 11, main text, are in these terms. In the concentration process, samples were reduced from 200 liters to an assay volume of 10 mL. The ANOVA and the standard errors are the same in either basis, as a constant factor of 20,000 (4.302 log-units) is involved in converting from one base to the other.

TABLE C-3. INTERMEDIATE ANALYSIS RESULTS CITED IN MAIN TEXT: ESCHERICHIA COLI

(a) Log Means and Standard Errors: Sample Point, Pooled and Flow-Partitioned

Sample Point	Pooled	Flow Rate, gpm		
		30	35	40
		Log Mean ± Standard Error[a]		
GS1	2.2775±0.1052(36)	2.2661±0.1402(12)	2.4087±0.2044(12)	2.1575±0.2033(12)
GS2	1.2943±0.1037(34)	1.3951±0.1433(12)	1.2797±0.2476(11)	1.1990±0.1472(11)
GS3	1.1174±0.1312(33)	1.2208±0.1429(12)	0.9618±0.2083(12)	1.1870±0.3592(9)

(b) Paired Comparisons

Paired Comparisons	Log Mean ± Standard Error
GS1-GS2	1.0439±0.1347
GS1-GS3	1.1843±0.1329
GS2-GS3	0.1405±0.1366

a. Observations associated with each result shown in parentheses.

TABLE C-4. INTERMEDIATE ANALYSIS RESULTS CITED IN MAIN TEXT: LS_c STRAIN, POLIOVIRUS I

(a) Log Means[a] and Standard Errors: Sample Points, Pooled and Flow-Partitioned

Sample Point	Pooled	Flow Rate, gpm		
		30	35	40
		Log Mean ± Standard Error[b]		
GS1	1.4251±0.1735	1.4513±0.2627	1.4574±0.2948	1.3667±0.4504
GS2	0.8715±0.1573	1.0252±0.2599	0.7506±0.2361	0.8388±0.3925
GS3	0.7201±0.1488	0.5074±0.3230	0.7059±0.2064	0.9471±0.2649

(b) Paired Comparisons

Paired Comparison	Log Mean ± Standard Error
GS1-GS2	0.5536±0.1486
GS1-GS3	0.7050±0.1486
GS2-GS3	0.1514±0.1486

a. Based on estimated PFU/mL of PV in unconcentrated sample.
b. Each pooled result based on nine values; each flow rate on three.

The total count ANOVA was fully-balanced; the intermediate results appear in Table C-5. The flow effect is associated with means of the combinations (log assay GS1 + log assay GS2 + log assay GS3) at each flow rate. The observation is that this mean at 35 gpm was quite different from the other two mean combinations. This can be appreciated by summing the GS1, GS2, and GS3 log means at each flow rate in part a of Table C-5 and dividing by 3; 30 gpm = 3.3532, 35 gpm = 3.0285, 40 gpm = 3.2707. These combinations have dubious operational meaning; the strong FxS interaction indicates that the individual log means are of more importance than their sums.

The total enterics were not assayed until the second test day. While the multifactor analysis can accommodate missing values interspersed through a schedule, the validity of the ANOVA with a full day's data missing is open to question. The decision was made to consider only the two full blocks of data in the ANOVA. The intermediate results from the ANOVA appear in Table C-6.

TABLE C-5. INTERMEDIATE ANALYSIS RESULTS CITED IN MAIN TEXT: TOTAL COUNT

(a) Log Means and Standard Errors: Sample Point, Pooled and Flow-Partitioned

Sample Point	Pooled	Flow Rate, gpm		
		30	35	40
		Log Mean ± Standard Error[a]		
GS1	3.6719±0.0886	3.9668±0.0699	3.5003±0.1688	3.5487±0.1718
GS2	3.0797±0.1014	3.1058±0.1487	3.2435±0.2396	2.8892±0.1132
GS3	2.9009±0.1455	2.9873±0.1918	2.3417±0.2661	3.3737±0.2126

(b) Paired Comparisons

Paired Comparison				
GS1-GS2	0.5922±0.1445	0.8610±0.2503	0.2568±0.2503	0.6588±0.2503
GS1-GS3	0.7710±0.1445	0.9795±0.2503	1.1586±0.2503	0.1750±0.2503
GS2-GS3	0.1788±0.1445	0.1185±0.2503	0.9018±0.2503	-0.4838±0.2503

a. Each pooled result based on 36 observations, each flow rate on 12 observations.

TABLE C-6. INTERMEDIATE ANALYSIS RESULTS CITED IN MAIN TEXT: TOTAL ENTERICS

(a) Log Mean and Standard Errors: Sample Point, Pooled and Flow-Partitioned

Sample Point	Pooled	Flow Rate, gpm		
		30	35	40
		Log Mean ± Standard Error[a]		
GS1	2.9632±0.0966	3.2476±0.1864	2.6907±0.1228	2.9511±0.1416
GS2	2.1741±0.1230	2.3032±0.2786	2.2483±0.1616	1.9708±0.1907
GS3	2.0828±0.1478	1.9627±0.1901	2.0773±0.2487	2.2084±0.3363

(b) Paired Comparisons

Paired Comparisons	Log Mean ± Standard Errors
GS1-GS2	0.7891±0.1818
GS1-GS3	0.8804±0.1818
GS2-GS3	0.0913±0.1818

a. Each pooled result based on 24 observations; each flow rate on 8 observations.

APPENDIX D

MULTIMEDIA FILTER TRANSIENT RESPONSE TIME

This experiment was performed to determine the transient response of the multimedia filter to sudden changes in inlet water composition. Such changes occur at start-up and shut-down, and test procedures must account for the duration of this response.

The method used was based on application of a step change to water content at the filter inlet and observation of its propagation at the filter outlet. Referring to Figure 2, main text, raw water pump #1 was started and river water was allowed to flow through the system. Five pounds of table salt (2.2 kg) were added to 20 liters of tap water and the mixture was stirred. The resulting solution was added to the system using the Poly Feed line arrangement. TDS measurements were taken every 2 minutes on samples drawn from the GS1 tap and the GS2 tap. Sampling was alternated: GS1 tap was sampled every even minute, and GS2 tap every odd minute. At t = 21 minutes, the Poly Feed valve was closed and the response of the system to a step decrease was observed. The average flow rate during the test was 30.5 gpm.

The resulting TDS measurement record is graphically portrayed in Figure D-1. From this experiment, 10 minutes was judged as sufficient time to account for transient effects.

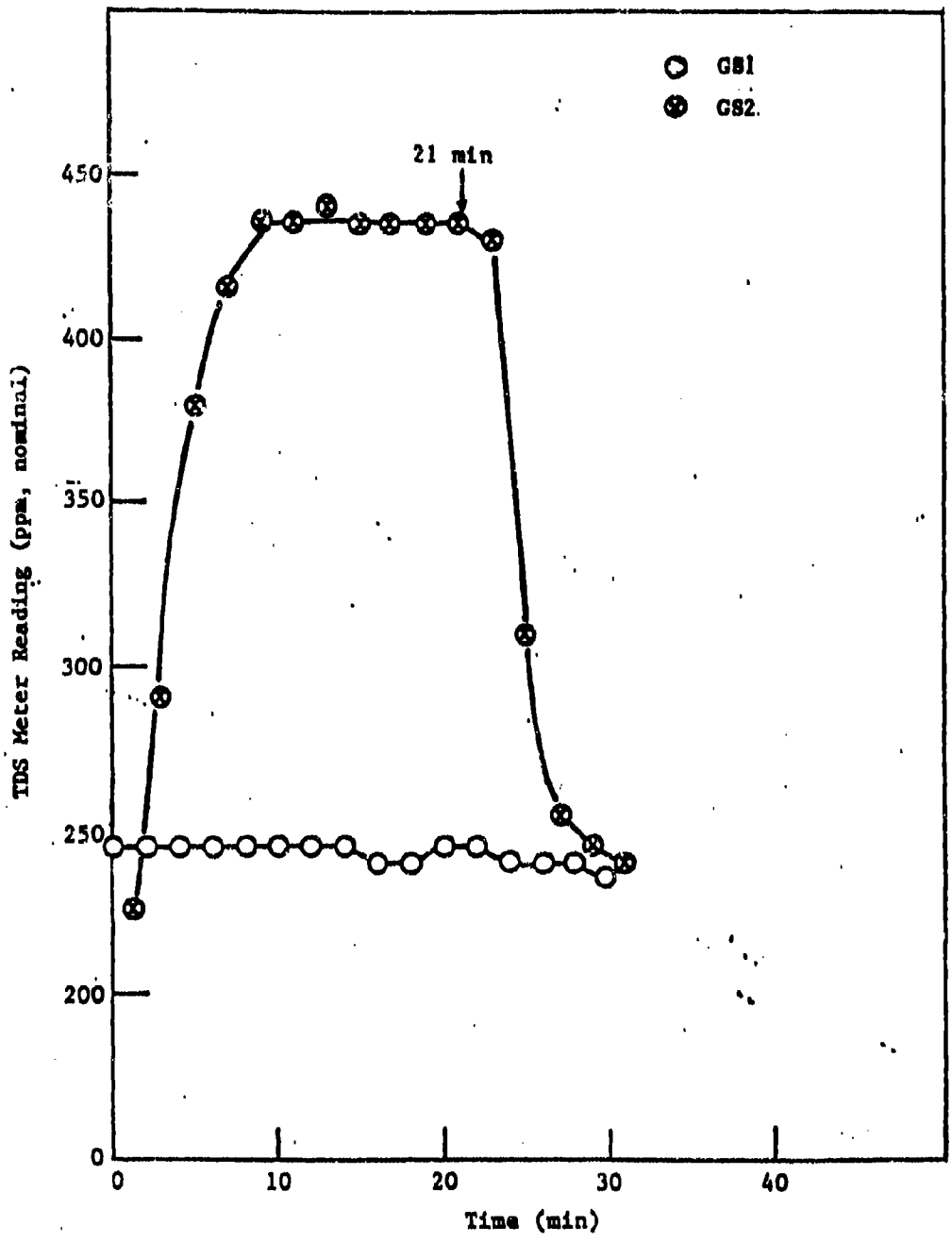

Figure D-1. Response of multimedia filter to step changes in inlet water TDS.

APPENDIX E

CALCULATION OF PHYSICAL AND CHEMICAL PROPERTIES: PHYSICAL AND CHEMICAL ANALYSIS

The total dissolved solids (TDS) content of a sample was based on the sample meter reading corrected by the meter reading estimate of a 200-mg/L sodium chloride solution. The correction was:

$$\text{TDS (reported)} = \text{TDS (sample)} \times \frac{267}{\text{TDS (200 mg/L NaCl)}}$$

The chlorine demand test was an attempt to determine the impact of the prefiltration system on substances that exert a chlorine demand. Knowledge of what this demand was would assist in a decision to prechlorinate or post-chlorinate in a prefiltration system configuration. The assay used was a simplification and modification of Standard Methods 410 B.* The changes were as follows: the titration solution was 1,000 ppm NaOCl, prepared by dilution of "Clorox" bleach (Clorox Corporation, San Diego, CA) with demand-free water (not standardized); 100 mL aliquots of sample used; two aliquots were titrated for each sample determination; and free available chlorine (FAC) was measured with a FACTS color comparator (LaMotte Chemical Company, Chestertown, MD).

The chlorine demand for each aliquot was computed in one of two ways. If the color developed after titration corresponded to a comparator reading,

chlorine demand = FAC (added by titration) - FAC (read)

If the color was intermediate between two comparator readings,

chlorine demand = FAC (added by titration) - average of bracketing FAC readings

The reported chlorine demand for a sample is the average of individual aliquot chlorine demands. A blank aliquot (no added FAC) was also assayed to assure that raw water had no FAC. In no instance did raw water exhibit FAC.

The TOC samples were pH-adjusted on site by the addition of 2 mL of 1 M sulfuric acid solution to 250 mL sample. Immediately prior to assay, the samples were sparged with air to remove inorganic carbon which was acid-converted to CO_2. Thus, the TOC reported is from what is strictly a total carbon analysis. A Beckman Model 915 Beckman Instrument Company TOC analyzer was used.

* Standard Methods for the Examination of Water and Wastewater, 14th ed. 1975. American Public Health Association, Washington, DC.

APPENDIX F

CHEMICAL, PHYSICAL, AND MICROBIOLOGICAL ASSAY DATA

The assay data are presented in seven tables:

Table	Contents
1	Turbidity Assays
2	Other Physical and Chemical Assays
3	*Bacillus globigii* Assays
4	*Escherichia coli* Assays
5	Poliovirus Assays
6	Total Counts Assays
7	Total Enterics Assays

The correspondence of days to statistical analysis structure is given in Table 5, main text. Times presented are the 24-hour clock time of sampling, or in the case of concentrated PV samples, the time at which concentration started.

Data that were not quantitative are prefixed with GT, LT, or MLT to indicate, respectively, a result considered greater, lower, or much lower than the number cited, which represents a detection limit. Samples in Table 5 are suffixed -C or -D to indicate that concentrated or direct samples are reported.

TABLE F-1. TURBIDITY ASSAYS

DATE	TIME	SAMPLE	TURB, NTU	DATE	TIME	SAMPLE	TURB, NTU
09/17	0815	GS1	3.5	10/09	0800	GS1	3.5
09/17	0826	GS2	1.1	10/09	0810	GS2	1.6
09/17	0828	GS3	0.74	10/09	0812	GS3	1.4
09/17	1112	GS1	3.5	10/09	1101	GS1	4.2
09/17	1122	GS2	0.55	10/09	1109	GS2	0.80
09/17	1124	GS3	0.40	10/09	1112	GS3	0.50
09/17	1411	GS1	3.7	10/09	1357	GS1	3.8
09/17	1421	GS2	0.48	10/09	1406	GS2	0.49
09/17	1422	GS3	0.37	10/09	1408	GS3	0.48
09/17	1649	GS1	4.0	10/09	1634	GS1	3.8
09/17	1700	GS2	0.41	10/09	1644	GS2	0.51
09/17	1701	GS3	0.33	10/09	1646	GS3	0.40
09/18	0802	GS1	6.0	10/10	0756	GS1	2.4
09/18	0811	GS2	0.78	10/10	0806	GS2	1.1
09/18	0812	GS3	0.55	10/10	0808	GS3	0.57
09/18	1055	GS1	5.9	10/10	1052	GS1	2.3
09/18	1105	GS2	0.51	10/10	1102	GS2	0.64
09/18	1106	GS3	0.39	10/10	1104	GS3	0.49
09/18	1355	GS1	5.5	10/10	1352	GS1	2.2
09/18	1405	GS2	0.43	10/10	1400	GS2	0.59
09/18	1407	GS3	0.32	10/10	1402	GS3	0.37
09/18	1633	GS1	5.6	10/10	1622	GS1	2.4
09/18	1643	GS2	0.36	10/10	1632	GS2	0.46
09/18	1644	GS3	0.30	10/10	1634	GS3	0.34
09/23	0805	GS1	4.7	10/15	0747	GS1	4.3
09/23	0815	GS2	1.1	10/15	0758	GS2	1.5
09/23	0816	GS3	1.2	10/15	0800	GS3	0.92
09/23	1100	GS1	3.9	10/15	1047	GS1	4.3
09/23	1110	GS2	0.64	10/15	1057	GS2	0.80
09/23	1114	GS3	0.66	10/15	1100	GS3	0.76
09/23	1358	GS1	3.8	10/15	1348	GS1	3.1
09/23	1407	GS2	0.60	10/15	1357	GS2	0.63
09/23	1408	GS3	0.58	10/15	1359	GS3	0.52
09/23	1637	GS1	4.0	10/15	1620	GS1	3.3
09/23	1647	GS2	0.59	10/15	1630	GS2	0.60
09/23	1649	GS3	0.54	10/15	1632	GS3	0.53

TABLE F-1 Continued

DATE	TIME	SAMPLE	TURB,NTU
10/22	0803	GS1	3.0
10/22	0814	GS2	1.9
10/22	0817	GS3	1.5
10/22	1103	GS1	3.1
10/22	1113	GS1	1.2
10/22	1116	GS3	0.84
10/22	1401	GS1	3.0
10/22	1410	GS2	0.87
10/22	1412	GS3	0.69
10/22	1633	GS1	3.1
10/22	1643	GS2	0.76
10/22	1647	GS3	0.64
10/23	0749	GS1	3.2
10/23	0801	GS2	1.2
10/23	0803	GS3	1.3
10/23	1047	GS1	2.9
10/23	1057	GS2	0.85
10/23	1100	GS3	0.77
10/23	1346	GS1	2.9
10/23	1355	GS2	0.82
10/23	1356	GS3	0.70
10/23	1619	GS1	3.2
10/23	1628	GS2	0.68
10/23	1630	GS3	0.62
10/29	0718	GS1	7.1
10/29	0732	GS2	1.8
10/29	0735	GS3	1.6
10/29	1011	GS1	6.7
10/29	1023	GS3	1.1
10/29	1311	GS1	5.1
10/29	1320	GS2	0.49
10/29	1322	GS3	0.46
10/29	1540	GS1	5.0
10/29	1552	GS2	0.40
10/29	1555	GS3	0.42

TABLE F-2. OTHER PHYSICAL AND CHEMICAL ASSAYS

DATE	TIME	SAMPLE	PH	TDS PPM	ALK MG/L, CACO3	TOC MG/L	CL DMD PPM
09/17	0815	GS1	7.9	278	120.4	5	1.2
09/17	0828	GS3	7.8	278	109.5	3	0.5
09/17	1112	GS1	8.0	275	116.8	6	1.0
09/17	1124	GS3	7.9	275	116.8	3	0.25
09/17	1411	GS1	8.4	270	116.8	10	0.0
09/17	1422	GS3	8.2	272	113.1	3	-0.5
09/17	1649	GS1	8.4	258	116.8	3	0.0
09/17	1701	GS3	8.3	255	113.1	3	0.0
09/18	0802	GS1	7.9	262	113.1	4	0.75
09/18	0812	GS3	7.9	267	120.4	5	0
09/18	1055	GS1	8.2	275	116.8	1	1
09/18	1106	GS3	8.1	265	113.1	2	1
09/18	1355	GS1	8.4	258	113.1	3	0.5
09/18	1407	GS3	8.3	252	113.1	6	0.5
09/18	1633	GS1	8.4	322	113.1	3	1.15
09/18	1644	GS3	8.3	305	113.1	4	0
09/23	0805	GS1	7.8	288	120.4	3	0.25
09/23	0816	GS3	7.8	286	120.4	4	-0.25
09/23	1100	GS1	7.8	267	120.4	5	1
09/23	1114	GS3	8.0	262	116.8	0	0.5
09/23	1358	GS1	8.4	262	113.1	4	1
09/23	1408	GS3	8.2	267	113.1	2	0
09/23	1637	GS1	8.4	260	116.8	0	1
09/23	1649	GS3	8.4	260	113.1	4	0
10/09	0800	GS1	7.9	274	153.9	5	1.1
10/09	0812	GS3	7.9	280	153.9	5	0.5
10/09	1101	GS1	8.1	286	173.8	2	1.45
10/09	1112	GS3	8.1	299	171.3	2	1.15
10/09	1357	GS1	8.4	267	158.9	4	1.3
10/09	1408	GS3	8.4	277	158.9	2	1.0
10/09	1634	GS1	8.5	260	158.9	3	1.3
10/09	1646	GS3	8.5	257	156.4	1	1.0
10/10	0756	GS1	8.1	282	158.9	6	1.7
10/10	0808	GS3	8.1	289	158.9	5	1.6
10/10	1052	GS1	7.9	267	148.9	6	1.8
10/10	1104	GS3	7.8	272	151.4	5	1.45
10/10	1352	GS1	7.8	270	151.4	12	1.0
10/10	1402	GS3	7.9	276	153.9	4	1.3
10/10	1622	GS1	7.8	294	153.9	4	1.7
10/10	1634	GS3	8.0	296	158.9	4	1.0
10/15	0747	GS1	7.8	289	148.9	4	1.15
10/15	0800	GS3	7.7	291	148.9	5	0.5
10/15	1047	GS1	7.8	300	158.9	4	1.45
10/15	1100	GS3	7.8	300	158.9	4	1.15
10/15	1348	GS1	8.0	278	158.9	4	1.6
10/15	1359	GS3	8.0	295	156.4	4	1.3
10/15	1620	GS1	8.1	272	156.4	4	1.3
10/15	1632	GS3	8.1	276	156.4	3	1.15

TABLE F-2 Continued

DATE	TIME	SAMPLE	PH	TDS PPM	ALK MG/L, CACO3	TOC MG/L	CL IMD PPM
10/22	0803	GS1	7.7	282	171.3	10	2.0
10/22	0817	GS3	7.7	275	166.3	8	1.1
10/22	1103	GS1	7.7	305	166.3	7	1.5
10/22	1116	GS3	7.6	322	158.9	8	1.15
10/22	1401	GS1	7.7	285	161.4	6	1.55
10/22	1412	GS3	7.8	278	161.4	6	1.0
10/22	1633	GS1	8.0	286	158.9	6	>2
10/22	1647	GS3	8.1	282	158.9	9	1.0
10/23	0749	GS1	7.7	309	156.4	4	1.8
10/23	0803	GS3	7.5	307	158.9	4	1.1
10/23	1047	GS1	7.5	293	158.9	4	1.65
10/23	1100	GS3	7.7	319	158.9	5	1.0
10/23	1346	GS1	8.0	304	163.8	3	1.7
10/23	1356	GS3	7.8	304	163.8	3	1.3
10/23	1619	GS1	7.8	285	158.9	4	1.95
10/23	1630	GS3	7.7	290	161.4	2	0.5
10/29	0718	GS1	7.6	>398	213.5	6	3
10/29	0735	GS3	7.8	>398	203.6	5	2.3
10/29	1011	GS1	7.9	>424	213.5	3	2
10/29	1023	GS3	7.8	396	213.5	3	2
10/29	1311	GS1	8.0	>445	228.4	3	2
10/29	1322	GS3	7.9	>445	228.4	2	0.5
10/29	1540	GS1	8.0	>452	248.2	3	2
10/29	1555	GS3	8.0	>452	243.3	2	1.15

TABLE F-3. BACILLUS GLOBIGII ASSAYS

DATE	TIME	SAMPLE	CFU/ML	DATE	TIME	SAMPLE	CFU/ML
09/17	0813	SEED	2.6E6	10/09	0755	SEED	7.18E6
09/17	0815	GS1	16.9E2	10/09	0800	GS1	33.E2
09/17	0826	GS2	2.35E2	10/09	0810	GS2	1.4E2
09/17	0828	GS3	1.1E2	10/09	0812	GS3	0.89E2
09/17	1106	SEED	3.8E6	10/09	1056	SEED	6.87E6
09/17	1112	GS1	13.2E2	10/09	1101	GS1	35.6E2
09/17	1122	GS2	1.49E2	10/09	1109	GS2	0.16E2
09/17	1124	GS3	1.05E2	10/09	1112	GS3	0.5E2
09/17	1407	SEED	2.3E6	10/09	1355	SEED	7.54E6
09/17	1411	GS1	12.0E2	10/09	1357	GS1	72.8E2
09/17	1421	GS2	0.57E2	10/09	1406	GS2	0.06E2
09/17	1422	GS3	1.8E2	10/09	1408	GS3	0.06E2
09/17	1646	SEED	6.6E6	10/09	1630	SEED	3.68E6
09/17	1649	GS1	10.6E2	10/09	1634	GS1	26.5E2
09/17	1700	GS2	1.67E2	10/09	1644	GS2	0.17E2
09/17	1701	GS3	0.693E2	10/09	1646	GS3	0.34E2
09/18	0757	SEED	3.3E7	10/10	0748	SEED	1.07E7
09/18	0802	GS1	71.0E2	10/10	0756	GS1	98.5E2
09/18	0811	GS2	1.0E2	10/10	0806	GS2	1.1E2
09/18	0812	GS3	0.519E2	10/10	0808	GS3	0.57E2
09/18	1055	GS1	22.2E2	10/10	1050	SEED	1.05E7
09/18	1105	GS2	1.41E2	10/10	1052	GS1	121.0E2
09/18	1106	GS3	MLTE2	10/10	1102	GS2	0.39E2
09/18	1352	SEED	2.9E7	10/10	1104	GS3	0.17E2
09/18	1355	GS1	150.E2	10/10	1352	GS1	86.6E2
09/18	1405	GS2	44.7E2	10/10	1400	GS2	0.42E2
09/18	1407	GS3	6.63E2	10/10	1402	GS3	0.24E2
09/18	1631	SEED	4.3E6	10/10	1618	SEED	1.23E7
09/18	1633	GS1	30.E2	10/10	1622	GS1	109.5E2
09/18	1643	GS2	6.32E2	10/10	1632	GS2	2.39E2
09/18	1644	GS3	0.31E2	10/10	1634	GS3	0.38E2
09/23	0755	SEED	8.40E6	10/15	0743	SEED	0.16E7
09/23	0805	GS1	33.4E2	10/15	0747	GS1	87.5E2
09/23	0815	GS2	2.22E2	10/15	0758	GS2	2.40E2
09/23	0816	GS3	7.75E2	10/15	0800	GS3	1.45E2
09/23	1100	SEED	1.38E6	10/15	1047	GS1	27.1E2
09/23	1100	GS1	78.7E2	10/15	1057	GS2	1.74E2
09/23	1110	GS2	4.0E2	10/15	1100	GS3	0.35E2
09/23	1114	GS3	0.3E2	10/15	1345	SEED	1.13E7
09/23	1357	SEED	1.22E6	10/15	1348	GS1	29.5E2
09/23	1358	GS1	14.3E2	10/15	1357	GS2	0.10E2
09/23	1407	GS2	0.1E2	10/15	1359	GS3	0.20E2
09/23	1408	GS3	1.0E2	10/15	1620	GS1	36.74E2
09/23	1630	SEED	1.70E6	10/15	1630	GS2	0.10E2
09/23	1637	GS1	15.7E2	10/15	1632	GS3	0.17E2
09/23	1647	GS2	0.1E2				
09/23	1649	GS3	MLTE2				

TABLE F-3. Continued

DATE	TIME	SAMPLE	CFU/ML
10/22	0800	SEED	8.85E5
10/22	0803	GS1	12.7E2
10/22	0814	GS2	1.2E2
10/22	0817	GS3	1.47E2
10/22	1100	SEED	1.36E7
10/22	1103	GS1	10.8E2
10/22	1113	GS2	3.00E2
10/22	1116	GS3	0.24E2
10/22	1400	SEED	1.05E6
10/22	1401	GS1	7.1E2
10/22	1410	GS2	0.14E2
10/22	1412	GS3	0.31E2
10/22	1630	SEED	1.93E6
10/22	1633	GS1	5.32E2
10/22	1643	GS2	0.10E2
10/22	1647	GS3	0.30E2
10/22	1631	RW	0.17E2
10/23	0747	SEED	7.48E4
10/23	0749	GS1	7.54E2
10/23	0801	GS2	0.72E2
10/23	0803	GS3	0.59E2
10/23	1045	SEED	3.95E5
10/23	1047	GS1	3.18E2
10/23	1057	GS2	0.14E2
10/23	1100	GS3	0.42E2
10/23	1345	SEED	3.79E5
10/23	1346	GS1	2.20E2
10/23	1355	GS2	0.14E2
10/23	1356	GS3	0.08E2
10/23	1615	SEED	2.55E5
10/23	1619	GS1	4.34E2
10/23	1628	GS2	0.15E2
10/23	1630	GS3	0.35E2
10/29	0709	SEED	3.69E6
10/29	0718	GS1	4.90E2
10/29	0732	GS2	0.20E2
10/29	0735	GS3	0.32E2
10/29	1010	SEED	6.60E6
10/29	1011	GS1	19.44E2
10/29	1021	GS2	0.28E2
10/29	1023	GS3	0.40E2
10/29	1310	SEED	1.23E6
10/29	1311	GS1	20.00E2
10/29	1320	GS2	0.20E2
10/29	1322	GS3	0.10E2
10/29	1535	SEED	3.39E6
10/29	1540	GS1	17.66E2
10/29	1552	GS2	0.69E2
10/29	1555	GS3	0.60E2

TABLE F-4. ESCHERICHIA COLI ASSAYS

DATE	TIME	SAMPLE	CFU/ML	DATE	TIME	SAMPLE	CFU/ML
09/17	0814	RW	3.5E1	10/09	0800	GS1	2.62E2
09/17	0815	GS1	1.15E2	10/09	0810	GS2	0.1E2
09/17	0826	GS2	.185E2	10/09	0812	GS3	0.3E2
09/17	0828	GS3	.20E2	10/09	1101	GS1	1.2E2
09/17	1112	GS1	.648E2	10/09	1109	GS2	0.07E2
09/17	1122	GS2	.125E2	10/09	1112	GS3	0.05E2
09/17	1124	GS3	.164E2	10/09	1357	GS1	0.63E2
09/17	1411	GS1	.648E2	10/09	1406	GS2	0.03E2
09/17	1421	GS2	.053E2	10/09	1408	GS3	0.04E2
09/17	1422	GS3	.036E2	10/09	1634	GS1	1.24E2
09/17	1649	GS1	1.47E2	10/09	1644	GS2	0.05E2
09/17	1700	GS2	.125E2	10/09	1646	GS3	0.01E2
09/17	1701	GS3	.1095E2	10/10	0756	GS1	1.15E2
09/18	0800	RW	6.5E1	10/10	0806	GS2	4.47E2
09/18	0802	GS1	85.0E2	10/10	0808	GS3	1.92E2
09/18	0811	GS2	1.0E2	10/10	1052	GS1	1.38E2
09/18	0812	GS3	MLTE2	10/10	1102	GS2	0.07E2
09/18	1055	GS1	6.32E2	10/10	1104	GS3	0.05E2
09/18	1105	GS2	MLTE2	10/10	1352	GS1	0.97E2
09/18	1106	GS3	MLTE2	10/10	1400	GS2	0.40E2
09/18	1355	GS1	8.95E2	10/10	1402	GS3	0.04E2
09/18	1405	GS2	1.0E2	10/10	1622	GS1	15.49E2
09/18	1407	GS3	MLTE2	10/10	1632	GS2	0.19E2
09/18	1630	RW	5.5E2	10/10	1634	GS3	0.24E2
09/18	1633	GS1	10.95E2	10/15	0744	RW	0.80E2
09/18	1643	GS2	MLTE2	10/15	0747	GS1	2.05E2
09/18	1644	GS3	1.0E2	10/15	0758	GS2	0.46E2
09/23	0805	GS1	72.8E2	10/15	0800	GS3	0.28E2
09/23	0815	GS2	1.65E2	10/15	1047	GS1	0.20E2
09/23	0816	GS3	11.8E2	10/15	1057	GS2	0.02E2
09/23	1100	GS1	3.74E2	10/15	1100	GS3	0.20E2
09/23	1110	GS2	.245E2	10/15	1348	GS1	1.28E2
09/23	1114	GS3	3.46E2	10/15	1357	GS2	0.17E2
09/23	1358	GS1	2.55E2	10/15	1359	GS3	0.02E2
09/23	1407	GS2	.20E2	10/15	1617	RW	1.64E2
09/23	1408	GS3	MLTE2	10/15	1620	GS1	1.12E2
09/23	1637	GS1	3.16E2	10/15	1630	GS2	0.45E2
09/23	1647	GS2	.490E2	10/15	1632	GS3	0.24E2
09/23	1649	GS3	.40E2				

TABLE F-4 CONTINUED

DATE	TIME	SAMPLE	CFU/ML
10/22	0802	RW	0.81E2
10/22	0803	GS1	0.75E2
10/22	0814	GS2	0.25E2
10/22	0817	GS3	0.17E2
10/22	1103	GS1	2.72E2
10/22	1113	GS2	0.32E2
10/22	1116	GS3	0.10E2
10/22	1401	GS1	3.00E2
10/22	1410	GS2	2.19E2
10/22	1412	GS3	0.10E2
10/22	1631	RW	0.24E2
10/22	1633	GS1	0.42E2
10/22	1643	GS2	LT E1
10/22	1647	GS3	0.75E2
10/23	0748	RW	0.20E2
10/23	0749	GS1	1.20E2
10/23	0801	GS2	0.30E2
10/23	1047	GS1	0.14E2
10/23	1346	GS1	0.24E2
10/23	1355	GS2	0.11E2
10/23	1356	GS3	0.05E2
10/23	1617	RW	0.35E2
10/23	1619	GS1	0.34E2
10/23	1628	GS2	0.11E2
10/29	0715	RW	0.49E2
10/29	0718	GS1	0.46E2
10/29	0732	GS2	0.24E2
10/29	0735	GS3	0.17E2
10/29	1011	GS1	4.58E2
10/29	1021	GS2	0.44E2
10/29	1023	GS3	0.65E2
10/29	1311	GS1	6.48E2
10/29	1320	GS2	0.40E2
10/29	1322	GS3	0.24E2
10/29	1538	RW	3.16E2
10/29	1540	GS1	6.78E2
10/29	1552	GS2	0.34E2
10/29	1555	GS3	0.14E2

TABLE F-5. POLIOVIRUS ASSAYS

DATE	TIME	SAMPLE	PFU/ML	DATE	TIME	SAMPLE	PFU/ML
09/17	0815	GS1-D	1.69E2	10/09	0755	SEED	2.54E6
09/17	1106	SEED	2.07E5	10/09	0800	GS1-D	7.23E2
09/17	1112	GS1-D	1.20E2	10/09	1056	SEED	2.71E6
09/17	1122	GS2-C	45.8E0	10/09	1101	GS1-D	5.44E2
09/17	1124	GS3-C	14.3E0	10/09	1109	GS2-C	3.96E0
09/17	1407	SEED	1.91E5	10/09	1112	GS3-C	2.11E0
09/17	1411	GS1-C	8.45E0	10/09	1355	SEED	2.01E6
09/17	1646	SEED	2.44E5	10/09	1357	GS1-D	4.40E2
09/17	1649	GS1-D	1.20E2	10/09	1357	GS1-C	25.4E0
09/17	1700	GS2-C	1.84E0	10/09	1630	SEED	2.16E6
09/17	1701	GS3-C	1.41E0	10/09	1634	GS1-D	7.14E2
09/18	0757	SEED	6.66E5	10/09	1644	GS2-C	7.4E0
09/18	0802	GS1-D	1.53E2	10/09	1646	GS3-C	3.18E0
09/18	1052	SEED	6.28E5	10/10	0748	SEED	2.41E6
09/18	1055	GS1-D	1.83E2	10/10	0756	GS1-D	1.39E3
09/18	1105	GS2-C	11.6E0	10/10	1050	SEED	1.58E6
09/18	1106	GS3-C	8.25E0	10/10	1052	GS1-D	1.21E3
09/18	1352	SEED	6.57E5	10/10	1102	GS2-C	31.2E0
09/18	1355	GS1-D	1.89E2	10/10	1104	GS3-C	17.1E0
09/18	1355	GS1-C	84.5E0	10/10	1346	SEED	6.71E5
09/18	1631	SEED	3.67E5	10/10	1352	GS1-D	1.52E3
09/18	1633	GS1-D	4.27E2	10/10	1352	GS1-C	48.6E0
09/18	1643	GS2-C	8.35E0	10/10	1618	SEED	6.15E5
09/18	1644	GS3-C	5.35E0	10/10	1622	GS1-D	6.16E2
09/23	0755	SEED	3.30E6	10/10	1632	GS2-C	9.05E0
09/23	0805	GS1-D	1.77E3	10/10	1634	GS3-C	11.3E0
09/23	1100	SEED	4.48E6	10/15	0743	SEED	1.63E6
09/23	1100	GS1-D	1.19E3	10/15	0747	GS1-D	4.89E2
09/23	1110	GS2-C	62.5E0	10/15	1045	SEED	2.67E6
09/23	1114	GS3-C	35.2E0	10/15	1047	GS1-D	3.87E2
09/23	1357	SEED	5.53E6	10/15	1057	GS2-C	8.70E0
09/23	1358	GS1-D	1.72E3	10/15	1100	GS3-C	12.5E0
09/23	1358	GS1-C	134E0	10/15	1345	SEED	2.24E6
09/23	1630	SEED	2.60E6	10/15	1348	GS1-D	9.12E2
09/23	1637	GS1-D	1.65E3	10/15	1348	GS1-C	34.0E0
09/23	1647	GS2-C	9.35E0	10/15	1615	SEED	1.97E6
09/23	1649	GS3-C	5.55E0	10/15	1620	GS1-D	6.87E2
				10/15	1630	GS2-C	9.95E0
				10/15	1632	GS3-C	6.55E0

TABLE F-5 Continued

DATE	TIME	SAMPLE	PFU/ML
10/22	0800	SEED	4.75E5
10/22	0803	GS1-D	7.47E2
10/22	1100	SEED	4.78E5
10/22	1103	GS1-D	2.72E2
10/22	1113	GS2-C	16.9E0
10/22	1113	GS2-D	2.97E2
10/22	1116	GS3-C	2.05E0
10/22	1116	GS3-D	2.83E2
10/22	1400	SEED	1.58E6
10/22	1401	GS1-D	2.72E2
10/22	1401	GS1-C	8.15E0
10/22	1630	SEED	5.37E5
10/22	1633	GS1-D	2.24E2
10/22	1643	GS2-C	2.13E0
10/22	1643	GS2-D	1.59E2
10/22	1647	GS3-C	2.13E0
10/22	1647	GS3-D	2.03E2
10/23	0747	SEED	7.19E5
10/23	0749	GS1-D	1.11E3
10/23	1045	SEED	4.38E5
10/23	1047	GS1-D	1.12E3
10/23	1057	GS2-C	3.19E0
10/23	1100	GS3-C	10.0E0
10/23	1345	SEED	5.22E5
10/23	1346	GS1-D	6.72E2
10/23	1346	GS1-C	3.75E0
10/23	1615	SEED	8.63E5
10/23	1619	GS1-D	8.99E2
10/23	1628	GS2-C	3.02E0
10/23	1630	GS3-C	15.45E0
10/29	0709	SEED	2.12E6
10/29	0718	GS1-D	1.47E3
10/29	1010	SEED	2.01E6
10/29	1011	GS1-D	1.25E3
10/29	1021	GS2-D	4.05E2
10/29	1021	GS2-C	18.0E0
10/29	1023	GS3-D	3.25E2
10/29	1023	GS3-C	2.57E0
10/29	1310	SEED	2.10E6
10/29	1311	GS1-D	1.38E3
10/29	1311	GS1-C	55.0E0
10/29	1535	SEED	1.78E6
10/29	1540	GS1-D	8.27E2
10/29	1552	GS2-D	9.5E1
10/29	1552	GS2-C	150.4E0
10/29	1555	GS3-D	6.9E1
10/29	1555	GS3-C	0.700E0

TABLE F-6. TOTAL COUNTS ASSAYS

DATE	TIME	SAMPLE	CFU/ML	DATE	TIME	SAMPLE	CFU/ML
09/17	0814	RW	9.1E3	10/09	0800	GS1	67E2
09/17	0815	GS1	83.8E2	10/09	0810	GS2	7.4E2
09/17	0826	GS2	74.2E2	10/09	0812	GS3	17.2E2
09/17	0828	GS3	90E2	10/09	1101	GS1	59.1E2
09/17	1112	GS1	109.5E2	10/09	1109	GS2	6.44E2
09/17	1122	GS2	30E2	10/09	1112	GS3	7.9E2
09/17	1124	GS3	28.3E2	10/09	1357	GS1	92.2E2
09/17	1411	GS1	118.1E2	10/09	1406	GS2	4.02E2
09/17	1421	GS2	64.8E2	10/09	1408	GS3	6.51E1
09/17	1422	GS3	73.5E2	10/09	1634	GS1	50.5E2
09/17	1649	GS1	104.9E2	10/09	1644	GS2	6.24E2
09/17	1700	GS2	49.0E2	10/09	1646	GS3	10.2E2
09/17	1701	GS3	94.9E2	10/10	0756	GS1	134E2
09/18	0800	RW	1.2E4	10/10	0806	GS2	19.95E2
09/18	0802	GS1	225.6E2	10/10	0808	GS3	1.1E2
09/18	0811	GS2	46.0E2	10/10	1052	GS1	133.5E2
09/18	0812	GS3	13.7E2	10/10	1102	GS2	5.74E2
09/18	1055	GS1	98.8E2	10/10	1104	GS3	4.31E2
09/18	1105	GS2	343.2E2	10/10	1352	GS1	110.8E2
09/18	1106	GS3	MLTE2	10/10	1400	GS2	5.88E2
09/18	1355	GS1	185E2	10/10	1402	GS3	4.52E2
09/18	1405	GS2	557.0E2	10/10	1622	GS1	175.8E2
09/18	1407	GS3	46.8E2	10/10	1632	GS2	10.33E2
09/18	1630	RW	1.1E4	10/10	1634	GS3	7.22E2
09/18	1633	GS1	100.1E2	10/15	0744	RW	19.97E2
09/18	1643	GS2	87.6E2	10/15	0747	GS1	103.0E2
09/18	1644	GS3	0.7E2	10/15	0758	GS2	18.25E2
09/23	0805	GS1	242.9E2	10/15	0800	GS3	11.43E2
09/23	0815	GS2	22.3E2	10/15	1047	GS1	38.79E2
09/23	0816	GS3	154.0E2	10/15	1057	GS2	6.13E2
09/23	1100	GS1	131.4E2	10/15	1100	GS3	3.04E2
09/23	1110	GS2	70.2E2	10/15	1348	GS1	45.43E2
09/23	1114	GS3	686.0E2	10/15	1357	GS2	9.79E2
09/23	1358	GS1	63.4E2	10/15	1359	GS3	2.31E2
09/23	1407	GS2	7.89E2	10/15	1617	RW	9.42E2
09/23	1408	GS3	174.7E2	10/15	1620	GS1	47.69E2
09/23	1637	GS1	68.9E2	10/15	1630	GS2	1.05E2
09/23	1647	GS2	10.5E2	10/15	1632	GS3	0.89E2
09/23	1649	GS3	6.95E2				

TABLE F-6 Continued

DATE	TIME	SAMPLE	CFU/ML
10/22	0802	RW	3.43E2
10/22	0803	GS1	14.27E2
10/22	0814	GS2	5.9CE2
10/22	0817	GS3	7.80E2
10/22	1103	GS1	13..3E2
10/22	1113	GS2	9.80E2
10/22	1116	GS3	3.64E2
10/22	1401	GS1	19.63E2
10/22	1410	GS2	2.30E2
10/22	1412	GS3	0.90E2
10/22	1631	RW	7.73E2
10/22	1633	GS1	7.46E2
10/22	1643	GS2	7.10E2
10/22	1647	GS3	1.53E2
10/23	0748	RW	1.89E2
10/23	0749	GS1	10.78E2
10/23	0801	GS2	8.48E2
10/23	0803	GS3	86.46E2
10/23	1047	GS1	6.18E2
10/23	1057	GS2	4.80E2
10/23	1100	GS3	5.74E2
10/23	1346	GS1	3.54E2
10/23	1355	GS2	2.74E2
10/23	1356	GS3	3.42E2
10/23	1617	RW	2.05E2
10/23	1619	GS1	6.29E2
10/23	1628	GS2	2.74E2
10/23	1630	GS3	15.53E2
10/29	0715	RW	11.29E2
10/29	0718	GS1	22.98E2
10/29	0732	GS2	2.37E2
10/29	0735	GS3	2.20E2
10/29	1011	GS1	36.23E2
10/29	1021	GS2	10.29E2
10/29	1023	GS3	9.44E2
10/29	1311	GS1	62.50E2
10/29	1320	GS2	4.73E2
10/29	1322	GS3	3.39E2
10/29	1538	RW	33.94E2
10/29	1540	GS1	51.38E2
10/29	1552	GS2	3.28E2
10/29	1555	GS3	3.63E2

TABLE F-7. TOTAL ENTERICS ASSAYS

DATE	TIME	SAMPLE	CFU/ML	DATE	TIME	SAMPLE	CFU/ML
09/18	0802	GS1	90.0E2	10/09	0800	GS1	26.5E2
09/18	0811	GS2	8.49E2	10/09	0810	GS2	1.6E2
09/18	0812	GS3	0.2E3	10/09	0812	GS3	0.89E2
09/18	1055	GS1	89.2E2	10/09	1101	GS1	27.0E2
09/18	1105	GS2	1.77E2	10/09	1109	GS2	0.33E2
09/18	1106	GS3	MLTE2	10/09	1112	GS3	0.33E
09/18	1355	GS1	61.9E2	10/09	1357	GS1	14.6E2
09/18	1405	GS2	96.3E2	10/09	1406	GS2	0.17E2
09/18	1407	GS3	18.9E2	10/09	1408	GS3	0.23E2
09/18	1633	GS1	35.2E2	10/09	1634	GS1	15.6E2
09/18	1643	GS2	32.5E2	10/09	1644	GS2	0.27E2
09/18	1644	GS3	26.0E2	10/09	1646	GS3	0.22E2
09/23	0805	GS1	111.9E2	10/10	0756	GS1	3.98E2
09/23	0815	GS2	12.2E2	10/10	0806	GS2	110.0E2
09/23	0816	GS3	85.2E2	10/10	0808	GS3	0.33E2
09/23	1100	GS1	81.6E2	10/10	1052	GS1	28.32E2
09/23	1110	GS2	11.1E2	10/10	1102	GS2	0.64E2
09/23	1114	GS3	763.9E2	10/10	1104	GS3	0.36E2
09/23	1358	GS1	14.1E2	10/10	1352	GS1	3.91E2
09/23	1407	GS2	1.45E2	10/10	1400	GS2	0.35E2
09/23	1408	GS3	120.9E2	10/10	1402	GS3	0.24E2
09/23	1637	GS1	81.1E2	10/10	1622	GS1	34.87E2
09/23	1647	GS2	3.95E2	10/10	1632	GS2	0.58E2
09/23	1649	GS3	6.8E2	10/10	1634	GS3	8.98E2
				10/15	0744	RW	2.14E2
				10/15	0747	GS1	3.39E2
				10/15	0758	GS2	5.39E2
				10/15	0800	GS3	4.88E2
				10/15	1047	GS1	2.04E2
				10/15	1057	GS2	0.32E2
				10/15	1100	GS3	2.25E2
				10/15	1348	GS1	3.39E2
				10/15	1357	GS2	4.88E2
				10/15	1359	GS3	0.07E2
				10/15	1617	RW	7.64E2
				10/15	1620	GS1	3.89E2
				10/15	1630	GS2	0.50E2
				10/15	1632	GS3	0.65E2

TABLE F-7 Continued

DATE	TIME	SAMPLE	CFU/ML
10/22	0802	RW	3.83E2
10/22	0803	GS1	10.18E2
10/22	0814	GS2	3.35E2
10/22	0817	GS3	14.95E2
10/22	1103	GS1	4.01E2
10/22	1113	GS2	1.91E2
10/22	1116	GS3	0.52E2
10/22	1401	GS1	25.40E2
10/22	1410	GS2	1.13E2
10/22	1412	GS3	0.65E2
10/22	1631	RW	2.28E2
10/22	1633	GS1	3.55E2
10/22	1643	GS2	3.18E2
10/22	1647	GS3	1.65E2
10/23	0748	RW	1.89E2
10/23	0749	GS1	3.64E2
10/23	0801	GS2	1.43E2
10/23	0803	GS3	73.86E2
10/23	1047	GS1	4.10E2
10/23	1057	GS2	5.40E2
10/23	1100	GS3	4.09E2
10/23	1346	GS1	2.67E2
10/23	1355	GS2	0.90E2
10/23	1356	GS3	0.53E2
10/23	1617	RW	2.12E2
10/23	1619	GS1	6.25E2
10/23	1628	GS2	3.48E2
10/23	1630	GS3	19.49E2
10/29	0715	RW	4.87E2
10/29	0718	GS1	4.69E2
10/29	0732	GS2	1.24E2
10/29	0735	GS3	0.66E2
10/29	1011	GS1	53.83E2
10/29	1021	GS2	4.12E2
10/29	1023	GS3	3.35E2
10/29	1311	GS1	48.96E2
10/29	1320	GS2	1.33E2
10/29	1322	GS3	1.05E2
10/29	1538	RW	33.98E2
10/29	1540	GS1	50.35E2
10/29	1552	GS2	2.84E2
10/29	1555	GS3	0.84E2

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